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(54) **PRODUCTION OF TRANSGENIC PLANTS  
WITH INCREASED SEED YIELD**

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(52) **U.S. Cl.** ..... **800/278; 800/298; 800/295;**  
**800/287; 800/267; 435/468**

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See application file for complete search history.

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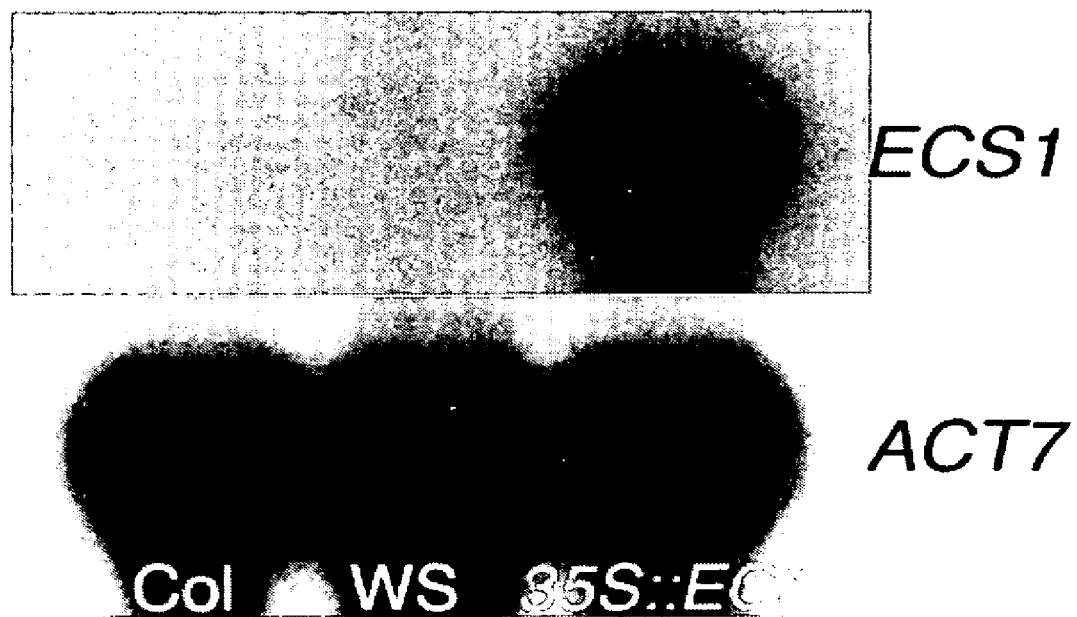
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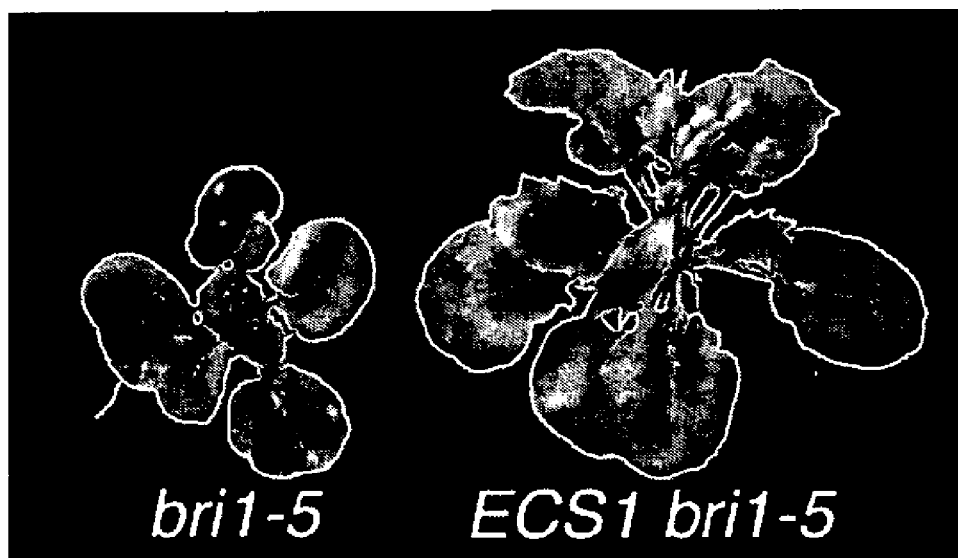
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(57) **ABSTRACT**

The invention provides methods of producing plants with  
increased seed production and transgenic plants with  
increased seed yields produced by said methods.



**FIG. 1**



**FIG. 2**

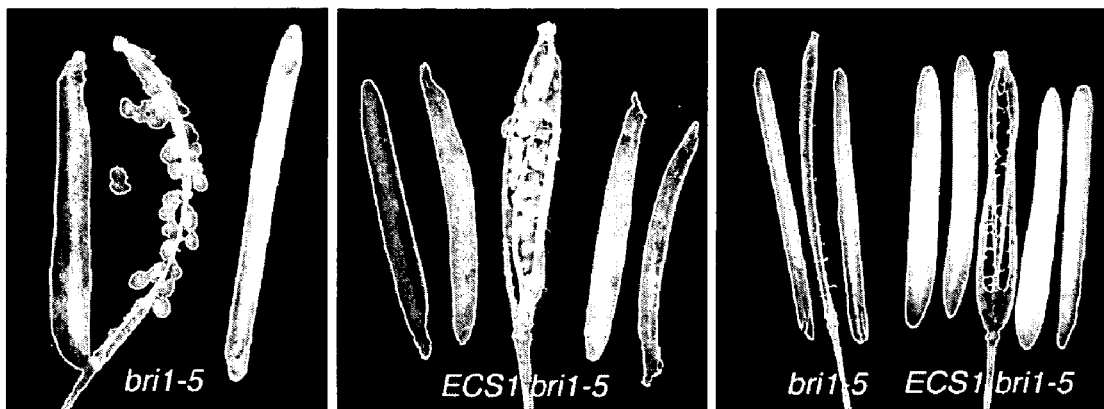


FIG. 3

ECS1	MARTHL-LFLLEV-LLSLA-----TSSTSTKEQEEEDRIKALPGQP-KVGFSGFSGYVTNESHGRSLFYWLTESS-SHSPHTKPL	76
BRS1	MARTHF-IFLLLVALLSTT-----FPSSSSREQEKDRIKALPGQP-KVAFSGYSGYVNVQSHGRALFYWLTESS-SPSPHTKPL	78
Homolog1	N-----KALPGQP-QVGFSGFSGYVTNESHGRSLFYWLTESS-SSHTKPL	46
Homolog2	MAMAKLAIPTTLMAILVMTSQGRIPTEGGEKEAEADRTSLPGQP-NVTFEQFSGYVTVDKLSGRSLFYWLTESS-DL-PLSKPL	82
Homolog3	MARL-LLLFFFLILLHYSCSR-----HEQEKDRIFHLPGEPNQVSFHFSGYITVNESAGRALFYWLTESSPPGENPESKPL	77
Homolog4	MDYS-FLLLILLLTI-STSCCAA--PSSVVEQLRDRI-SNLPGQPSNVDFAQYSGYVTVHEERGRALFYWLTESSPLARDPKSRPL	81
ECS1	LLWLNGGPGCSSIAYGASEEIGPFRISKTGCLNLYLNNSWNTEANLLFLESPVGVGFSYNTISSDFEESGDERTAQENLIFLISW	161
BRS1	LLWLNGGPGCSSIAYGASEEIGPFRINKTGSNLYLNKFAWNKDANLLFLESPAGVGFSYNTISSDLKDSGDERTAQENLIFLIKW	163
Homolog1	LLWLNGGPGCSSIAYGASEEIGPFRINKTGSNLYLNKFTWNTKANLLFLESPAGVGFSYNTISSDLKDSGDERTAQENLIFLIKW	131
Homolog2	VLWLNGGPGCSSIAYGASEEIGPFRISKGSGGLYLNKFAWNISNLLFLEAPAGVGFSYNTNRSSDLFNTGDRRTAKDSLQFLIQW	167
Homolog3	VLWLNGGPGCSSIAYGASEEIGPFRINPDQKTLYHNPYSWNKLANLLFLESPAGVGFSYNTISSDLVTAGDORTAFDAYVFLVKW	162
Homolog4	VLWLNGGPGCSSIAYGASEEIGPFRVGSQDKTILSKLYAWNKLANLLFLESPAGVGFSYNTISSDLVTTGDORTAEDSYFLVNW	166
ECS1	MSRFPQYRDFYIAGESYAGHYVPOLAKKIHEYNNAKKN-PVNLKGFVGNPEMDKNNDRLGILTYWWSHAMISDASYNRIK	245
BRS1	LSRFPQYRDFYIAGESYAGHYVPOLAKKINDYNKAFSK-PI NLKGFVGNNAVTDNQYDSTIGILTYWWSHAMISDKSYKSLK	247
Homolog1	MSRFPQYRDFYIAGESYAGHYVPOLAKKILYLNKAFNNPI NLKGFVGNNGDMDSHYDRLGAAWWSHAMISDKTYKSLK	216
Homolog2	LHREFQYHRELYIAGESYAGHYVPOLAKELMNYNKK--RSKNPLNLKGFVGNNAVTDNHYDNLGTVSYWWSHAMISDRIVHQLIS	250
Homolog3	FERFPQYKHEFYIAGESYAGHYVPOLSDIYVYK-----RN-PA NLKGFVGNNAVTDYHDYVGLFEYWAHGLISDLTYHNLRI	242
Homolog4	FERFPQYKHEFYIAGESYAGHYVPOLSKLVHERNKGFKN-PA NLKGFVGNNAVTDYHDYVGLFEYWAHGLISDSTVHQLKT	250
ECS1	NCDF-IADRESKECDAL-VYAAADFGDIQOYSIYTPKCYPPQDOT---NOTKFEOMMON-HITKRFLEDQY--DPCTENYAEIY	322
BRS1	YCNF-IYEBVSDDDQNAVNYANNHEFGDIQOYSIYTPCYAADQKK---NTIGFVRMKN-TLLRRLVSGY--DPCTESYAEKY	325
Homolog1	NCSE-IADKTDDKNWAL-VFYREFGKVNYSIYSPCN---HOT---NOTKFE---L-HG--GILLVEEYEDPCTESYAEIY	285
Homolog2	TCDF-SSRQKEIDLETLYSAMQEFGNIQOYNIAAPCNKSSDGGGSYNGSSGRRSRALPLPHSVLRKISGYDPCTERYAEIY	334
Homolog3	TCESGSEHPSKCIKAME-ANDLEQGNIDPYSIYVIL-----KKEAAALRSFESRVRHPWMMR-----AYDPCTEKYSGMY	314
Homolog4	AGYSYSQHPMSQCNVALR-NAELEQGNIDPYSIYFLKPC-----NS-TVALK-RFLKGRYPWMSR-----AYDPCTERYSNVY	320
ECS1	YNRPDORAMHANHAIPIYKWLACSDVLIKIL-KDSDKIMLPIYKELIAGLRIVWVYSGDIDSVIPVITATRYSLGKILNVPKTR	407
BRS1	JNRPDORAMHANVIGIYKWLACSDVLIKIL-KDSDKIMLPIYKELIAGLRIVWVYSGDIDSVIPVITATRFELSHILNVPKTR	408
Homolog1	YNRPDORAMHANLSIPIYKWLACNVVNNN--WKDSFSLMPLIYKELIAGLRIVWVYSGDIDAVVPVITATRLALSKILNVPKTR	368
Homolog2	YNRPDORAMHANTIKIPIYKWLACSEVL--NRNWDIDSVIPYDEMAGGLRIVWVYSGDIDSVVPVITATRYSLARLSLSTKLPL	417
Homolog3	FNSPEVQAMHANIIGLAIPWKGSQIVGEK--WADSPSLMPLIYKELIAGLRIVWVYSGDIDSVVPVITATRYSLRALKLQPLSK	397
Homolog4	FNRPDVQKALHANVIRLSIPWKACSDIIGESY--WDDSPSLMPLIYKELIAGLRIVWVYSGDIDAVVPVITATRYSDVALKELATILN	403
ECS1	WYPWYSGNQVGGRTVEYEGLTFTYIVRAGAGHEVPLFEPQDASALLRSFLAGNELSRSY	464
BRS1	WYPWYTDNQVGGWIEVYKGLTFPIIVRAGAGHEVPLFEPKRALILERSFLAGKELPRSY	465
Homolog1	WYPWYSIKQVGGWIEVYEGLTFTPIIVRAGAGHEVPLFQERALTILNSTLAGKELPRSY	425
Homolog2	WYPWYKKQVGGWIEVYEGLTFTPIIVRAGAGHEVPLFKPRAAELFKYFLRQKPLPKA	473
Homolog3	WYPWYDQGVGGWSQVYKGLTILVTHGAGAGHEVPLERRRAFLFLQSFQDNKPLPKA	452
Homolog4	WYPWYDHGKVGGSQVYKGLTILVTVAGAGHEVPLHRRRQAFILFIRSTLESKPMPMT	459

FIG. 4

ECS1	MARTH---	LLF-----	LLFVLLS---	-----	ATSSSTSTKE--	QEDRIKALPGQP--	KVGF	SQFSGYVT	50													
Barley ECS1	MTTIT---	RRLPPAPAAAAV	LLAALTC	LLRPA	AAVAGGH-----	AAADRI	VRLPGQP--	EVDF	DMYSGYIT	62												
Rice ECS1	MAAAA---	VLL-----	AAI	LLA--LSP	LPMSLSAGGGGGG	GTAE--	AAADRI	TALPGQP--	RVNF	SMYSGYVT	61											
Rice ECS1/H1	MKVQTS	SPCLLL	LLGSLALVT	LT	LCGPAASARPET	GSLSA	LAAMEL	DEL	DRVMS	LPGQ	AYSPE	ROYSGYVT	75									
Rice ECS1/H2	M-----	-----	-----	-----	-----	ELDEL	DRVMS	LPGQ	AYSPE	ROYSGYVT	30											
ECS1	VNESHGRSLFYWLT	ESSSHSPHTKPI	LLWNGGPGCSSI	AYGASEEL	GPFRISKTCN	LYLNF	NFSWN	TEANLLFL	125													
Barley ECS1	VDEAAGRSIFYL	QAP-EEA	OPAPLVL	WNGGPGCSSY	AYGASEEL	CAFRVMP	RGAGLVN	EYRWN	KVANYLFL	136												
Rice ECS1	VDAAAGRALFYWL	CAA-D-PAS	APLVL	WNGGPGCSSY	GYGASEEL	CAFRIN	PDGRSLYL	NYPWN	RVANMLFL	134												
Rice ECS1/H1	TDEYLGKALFYWFL	EAT-DKP	DEKPLVL	WNGGPGCSSI	GFQAAQL	GPFL	VKKDVA	EELN	PYAWN	QVANLLFL	149											
Rice ECS1/H2	TDEYLGKALFYWFL	EAT-DKP	DEKPLVL	WNGGPGCSSI	GFQAAQL	GPFL	VKKDVA	EELN	PYAWN	QVANLLFL	104											
ECS1	ESPVGVGFSYNTNTSS	DFEESGDERTAOEN	LIFLISWMS	RFPQYRYRDFY	IVGESYAGHYVP	OLAOKI	HEYNNAY-	199														
Barley ECS1	DSPAGVGFSYNTNTSS	DIYTS	GDNRTAHDSYAF	LAAWFER	FPHYKYRE	FVAGESYAGHYVP	ELSOLV	HRS	GN---	208												
Rice ECS1	DSPAGVGFSYNTNTSS	DLFTAGDN	KTAHDSYAF	LVNWLER	FPOYKYRDFY	IAGESYAGHYVP	QLSOLV	YRN	NKDV-	208												
Rice ECS1/H1	DSPAGVGFSYNTNTS	FGKDP	PGDNSTAYG	SYTFLIRWF	QRFQHKMK	EYIAGESYAGHYVP	QLANV	VDQ	NKI	AP	224											
Rice ECS1/H2	DSPAGVGFSYNTNTS	FGKDP	PGDNSTAYG	SYTFLIRWF	QRFQHKMK	EYIAGESYAGHYVP	QLANV	VDQ	NKI	AP	179											
ECS1	KNPVNLKGF	MVGNPEMDKNN	DLGTITVWWS	HAMISDASYNRI	LKNC	DFTA--	DRFS	KECDS	AIYVA	AADFG	DI	273										
Barley ECS1	--PVLNLKGF	MVGNGLI	DDYHDYVGT	FEFWNNH	GIVSDDTYR	LKDA	CHDSFI	HPSPAC	DAAT	DVAT	AEQ	NI	281									
Rice ECS1	EKPI	LNFKGF	MVGNNAVI	DDYHDYVGT	FEFWNNH	GILSDDTY	QKLQVAC	DFESSA	HAHSEAC	NKIYEVA	EAQ	NI	283									
Rice ECS1/H1	KENY	NLKGIM	GNAYMDG	DTDLG	IVDSAWH	HALISDK	LYSDFQK	FCNF--	SLVDL	SKECNA	AI	DQFNALYSI	DI	298								
Rice ECS1/H2	KENY	NLKGIM	GNAYMDG	DTDLG	IVDSAWH	HALISDK	LYSDFQK	FCNF--	SLVDL	SKECNA	AI	DQFNALYSI	DI	253								
ECS1	OYSLYTPKCVPP	ODOTNDIKFFQ	MMOMHTTKRFL	EDQYD	PCTENYAEI	YNNRPE	VORAMHANHT-	AI	PKWTACS	347												
Barley ECS1	MYSLYTPVCN	ISS--SSSS	SLSRRTGRYP	MTGS	YDCTERYSTAY	NNRD	VO	TALHAN	VTG	MNYT	WNC	354										
Rice ECS1	AYSLYTP	PTCK-----	KLS	ELKRR	LIRGNSP	MLPRGYD	PCLEKYST	KYNNL	DEVO	KAFHAN	YIG--	IPYAW	ITCS	350								
Rice ECS1/H1	IYSLYTP	PRCEL	GYPNFNS	SFAA	I	GRTSSR---	I	PMGYD	PCSQTYATE	YFNK	DVOKAL	HANI	PGA--	YS--LCH	356							
Rice ECS1/H2	IYSLYTP	PRCEL	GYPNFNS	SFAA	I	GRTSSR---	I	PMGYD	PCSQTYATE	YFNK	DVOKAL	HANI	PGA--	YS--LCH	321							
ECS1	DSVFNNWNW	SDNS	MLPIYKEL	I	AAGLR	WVYS	GD	TSV	IPVTAT	RYSLG	KLNL	RVKTR	WYFWYS	Q--NOVGGRT	421							
Barley ECS1	DTIN	TH--W	DA	PR	MLPIYREL	I	AAGLR	WVYS	GD	TSV	IPVTAT	RYSLG	KLNL	RVKTR	WYFWYS	Q--NOVGGRT	427					
Rice ECS1	DDLY	Y--W	DA	PR	MLPIYREL	I	AAGLR	WVYS	GD	TSV	IPVTAT	RYSLG	KLNL	RVKTR	WYFWYS	Q--NOVGGRT	422					
Rice ECS1/H1	NSI--	N	RAW	DS	DMTVL	PIV	KKLT	QSGL	R	W	YS	GD	TARI	PTT	STRY	TLK	KLGLPI	KED	WS	PWFH	--KQVGGWS	438
Rice ECS1/H2	NSI--	N	RAW	DS	DMTVL	PIV	KKLT	QSGL	R	W	YS	GD	TARI	PTT	STRY	TLK	KLGLPI	KED	WS	PWFH	--KQVGGWS	393
ECS1	EVYEG	LTFVT	VRGAGHE	VPFFQ	PQSALI	LLRS	FLAGNELS-	RSY	464													
Barley ECS1	QVYQ	GLTLV	VRGAGHE	VP	LHRR	PRQAL	LFQ	FLQ	GKMP	GR--	TTN	VTVA	476									
Rice ECS1	QVYQ	GLTLV	VRGAGHE	VP	LHRR	PRQAL	LFQ	FLQ	GKMP	GR--	TTN	VTVA	490									
Rice ECS1/H1	VVFD	GLTFVT	VRGAGH	MVPSI	MPE	QALE	LF	KY	FLANQN	PSKPF	482											
Rice ECS1/H2	VVFD	GLTFVT	VRGAGH	MVPSI	MPE	QALE	LF	KY	FLANQN	PSKPF	437											

FIG. 5

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## PRODUCTION OF TRANSGENIC PLANTS WITH INCREASED SEED YIELD

This application claims the priority of U.S. Provisional Patent Appl. Ser. No. 60/599,378, filed Aug. 6, 2004, the entire disclosure of which is specifically incorporated herein by reference.

### BACKGROUND OF THE INVENTION

#### 1. Field of the Invention

The present invention relates generally to the field of molecular biology. More specifically, the invention relates to methods and compositions for increasing plant seed yield.

#### 2. Description of the Related Art

BRS1 is a secreted serine carboxypeptidase that is implicated in an early step in brassinosteroid signaling, probably by taking part in the proteolytic processing of a protein involved in activating the BRI1 receptor (Li et al., 2001). The protease activity of BRS1 is required for its function in suppressing the phenotypes of a weak BRI1 allele, *bri1-5*. BRI1 is a member of a serine carboxypeptidase gene family in *Arabidopsis*. The fact that a loss-of-function allele of BRS1 does not show any significant phenotypes suggests that there is functional redundancy among the family members.

It has been shown that BRS1 overexpression suppresses multiple *bri1* defects, suggesting BRS1 might play an important role in an early stage of the BRI1 signaling pathway (Li et al., 2001). The presence of an N-terminal signal peptide in BRS1 predicts that the protein should enter the secretory pathway. Sequence analysis failed to identify any obvious endoplasmic reticulum or Golgi apparatus retention sequences. Therefore, BRS1 may be a secreted protein. These observations are consistent with findings that BRS1 suppressed two extracellular domain mutants, *bri1-5* and *bri1-9*, but failed to suppress a loss-of-function cytoplasmic domain mutant *bri1-1* (Friedrichsen et al., 2000).

BRS1 shares homology with another serine carboxypeptidase II-like protein, designated ECS1. Like BRS1, ECS1 is predicted to have an N-terminal signal peptide and should be secreted. Based on its biochemical properties, yeast Kex1p is classified in the same carboxypeptidase group (carboxypeptidase D). In yeast, both Kex1p and Kex2p/kexin are required for the maturation of peptide hormones,  $\alpha$ -mating pheromone and K1 killer toxin, from their inactive precursors (Dmochowska et al., 1987; Fuller, 1989). Kex2p/kexin is a membrane bound endoprotease, which specifically cleaves on the carboxyl side of pairs of basic amino acids (e.g. KR↓ or RR↓). Kex2p related endoproteases are also known as subtilisin and furin (Barr, 1991). Following the action of Kex2p/Kexin, Kex1p selectively trims off the flanking amino acids from the C-terminus of processing intermediates.

There are numerous examples of the importance of carboxypeptidases in ligand processing in animals. For example, a mutation in carboxypeptidase E (CPE), a metallopeptidase, results in the fat mouse mutant (Naggert et al., 1995; Fricker and Leiter, 1999). CPE is widely distributed in brain, pituitary and other neuroendocrine tissues and is thought to be involved in the processing the precursors of neuroendocrine peptides (Naggert et al., 1995; Fricker and Leiter, 1999).

In addition to ligand processing, there are also examples of receptor proteolytic processing. One example of receptor processing is the insulin receptor. Both insulin and insulin receptor are synthesized as inactive precursors. Proinsulin and insulin proreceptors are processed by furin-like endoproteases in the trans Golgi network to form active molecules, which recognize and cleave at the carboxy terminal sites of

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dibasic amino acids. Proinsulin is processed at the C-termini of KR and KTRR sites. The insulin proreceptor is processed at the RKRR site (Barr, 19991).

In plants, there are a few reports concerning the processing of ligand-like peptides or receptor-like proteins. In response to wounding, tomato systemin is processed from its inactive form, preprosystemin (Schaller and Ryan, 1994). Also in tomato, a secreted leucine-rich repeat protein (LRP), which was thought to be involved in a plant defense response, is proteolytically processed during pathogenesis (Tornerio et al., 1996). It is not clear whether prosystemin is cleaved by a subtilisin-like endoprotease, but it has been found that systemin physically interacts with a subtilisin-like protein SPB50 (Schaller and Ryan, 1994). LRP is likely to be processed by a subtilisin/Kex2p-like endoprotease (Tornerio et al., 1996). Additionally, the functions of two *Arabidopsis* Kex2p-like genes have been determined: AIR3 is involved in the regulation of auxin-induced lateral root formation (Neuteboom et al., 1999) and SDD1 functions in guard cell development (Berger and Altmann, 2000).

The regulatory roles of serine carboxypeptidases in plants have not yet been investigated. Therefore, while the foregoing studies have further understanding of plant metabolism, a beneficial use for numerous serine carboxypeptidases and for ECS1 and its orthologs in particular has been lacking.

### SUMMARY OF THE INVENTION

In one aspect, the invention provides a transgenic plant expressing a selected DNA conferring increased seed production and/or yield to the plant relative to a second plant of the same genotype lacking the selected DNA. In certain embodiments of the invention, the selected DNA comprises the nucleic acid sequence of SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7; SEQ ID NO:9; SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30 or SEQ ID NO:32. In another embodiment, the selected DNA encodes a polypeptide selected from the group consisting of SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31 and SEQ ID NO:33. In still another embodiment, the selected DNA is further defined as hybridizing to the nucleic acid sequence of SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7; SEQ ID NO:9; SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30 or SEQ ID NO:32 under conditions of 5×SSC, 50% formamide and 42° C. In still another embodiment, the selected DNA is further defined as encoding a polypeptide comprising at least 90% amino acid identity to a sequence selected from the group consisting of SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31 and SEQ ID NO:33.

A transgenic plant provided by the invention may comprise a selected DNA operably linked to a heterologous promoter. Such a promoter may be, for example, a developmentally-regulated, organelle-specific, inducible, tissue-specific, constitutive, cell-specific, seed specific, or germination-specific promoter. In certain embodiments, the selected DNA further comprises at least one additional sequence chosen from the group consisting of: a regulatory sequence, a selectable or

screenable marker, a leader sequence and a terminator. The transgenic plant may be further defined as a monocotyledonous plant. Examples of such plants include wheat, maize, rye, rice, oat, barley, sorghum or millet. The plant may further be a dicotyledonous plant. Examples of such plants include tomato, potato, soybean, canola, alfalfa, pea or sunflower. The transgenic plant may further be defined as a progeny plant of any generation of an R<sub>0</sub> transgenic plant, wherein the transgenic plant has inherited the selected DNA from the R<sub>0</sub> transgenic plant.

The invention also provides parts of a transgenic plant of the invention. In one embodiment such a part is a seed, wherein the seed comprises the selected DNA. A cell of a plant of the invention is also provided. Such a cell may be defined as expressing a protein encoded by the selected DNA. The cell may have inherited the selected DNA from a progenitor of the cell, and may have been transformed with the selected DNA.

In another aspect, the invention provides a transformation construct comprising an isolated nucleic acid sequence encoding a polypeptide having at least 90% amino acid identity to a polypeptide selected from the group consisting of SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31 and SEQ ID NO:33, wherein the isolated nucleic acid sequence is operably linked to a heterologous promoter. The isolated nucleic acid sequence may be further defined as comprising the nucleic acid sequence of SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30 or SEQ ID NO:32. In further embodiments, the isolated nucleic acid sequence may encode a polypeptide selected from the group consisting of SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31 and SEQ ID NO:33. In still further embodiments, the isolated nucleic acid sequence may hybridize to the nucleic acid sequence of SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30 or SEQ ID NO:32 under conditions of 5×SSC, 50% formamide and 42° C. The heterologous promoter may, for example, be a developmentally-regulated, organelle-specific, inducible, tissue-specific, constitutive, cell-specific, seed specific, or germination-specific promoter. A nucleic acid provided by the invention may be defined, for example, as having at least 70%, 80%, 85%, 90%, 95%, 98%, 99% or more sequence identity to one or more nucleic acid sequence(s) selected from SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30 or SEQ ID NO:32.

In yet another aspect, the invention provides a method for increasing seed production and/or yield in a plant comprising introducing into the plant a nucleic acid sequence selected from the group consisting of: (a) the nucleic acid sequence of SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID

NO:30 or SEQ ID NO:32; (b) a nucleic acid sequence encoding the polypeptide of SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31 or SEQ ID NO:33; (c) a nucleic acid sequence defined as hybridizing to the nucleic acid sequence of SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7, SEQ ID NO:9, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20, SEQ ID NO:22, SEQ ID NO:24, SEQ ID NO:26, SEQ ID NO:28, SEQ ID NO:30 or SEQ ID NO:32 under conditions of 5×SSC, 50% formamide and 42° C.; and (d) a nucleic acid sequence encoding a polypeptide comprising at least 90% amino acid identity to the polypeptide sequence of SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8, SEQ ID NO:10, SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21, SEQ ID NO:23, SEQ ID NO:25, SEQ ID NO:27, SEQ ID NO:29, SEQ ID NO:31 or SEQ ID NO:33.

In a method of the invention, the isolated nucleic acid sequence may be defined as from a species selected from the group consisting of: *Arabidopsis thaliana*, barley, potato, rice, pea, tomato, wheat and alfalfa. In such a method the number of seed produced by the plant may be increased relative to a second plant of the same genotype lacking the isolated nucleic acid and/or the weight of seed produced by the plant may be increased relative to a second plant of the same genotype lacking the isolated nucleic acid. Introducing the isolated nucleic acid may comprise plant breeding and may comprise genetic transformation.

In still yet another aspect, the invention provides a method of making food for human or animal consumption comprising: (a) obtaining a plant of the invention; (b) growing the plant under plant growth conditions to produce plant tissue from the plant; and (c) preparing food for human or animal consumption from the plant tissue. In the method preparing food may comprise harvesting the plant tissue. The food may be starch, protein, meal, flour or grain.

In still yet another aspect, the invention provides a method of preparing seed comprising: (a) obtaining a plant of the invention; (b) growing the plant under plant growth conditions to produce seed; and (c) collecting seed produced by the plant.

The use of the word “a” or “an” when used in conjunction with the term “comprising” in the claims and/or the specification may mean “one,” but it is also consistent with and encompasses the meaning of “one or more,” “at least one,” and “one or more than one.”

Other objects, features and advantages of the present invention will become apparent from the following detailed description. It should be understood, however, that the detailed description and the specific examples, while indicating specific embodiments of the invention, are given by way of illustration only, since various changes and modifications within the spirit and scope of the invention will become apparent to those skilled in the art from this detailed description.

#### BRIEF DESCRIPTION OF THE DRAWINGS

The following drawings form part of the present specification and are included to further demonstrate certain aspects of the invention. The invention may be better understood by reference to one or more of these drawings in combination with the detailed description of specific embodiments presented herein:



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FIG. 1. Overexpression of ECS1 in wildtype *Arabidopsis*. Both Columbia and WS ecotypes express a very low level of ECS1, while the ECS1-overexpressing line (in WS background) has an elevated ECS1 expression level (top panel). ACT7 was used as a probe to show the sample equal loading of total RNA (bottom panel).

FIG. 2. Overexpression of ECS1 suppresses bri1-5 phenotypes. Rosette leaves in BRI1-5 are curled, while ECS1-overexpressing plants have expanded leaves. bri1-5 plants flower 7-10 days later than wildtype plants, while ECS1-overexpressing plants flower 5-7 days earlier than bri1-5 plant.

FIG. 3. Siliques of bri1-5 and ECS1 bri1-5. The left and center panels show the dissected green siliques with seeds attached. The right panel compares the siliques of bri1-5 and ECS1 bri1-5 after removal of the seeds. Four carpels in ECS1 bri1-5 contrast two carpels in bri1-5.

FIG. 4. Alignment of the predicted amino acid sequence of ECS1 (SEQ ID NO:2) with the predicted amino acid sequences of the five most related genes in *Arabidopsis thaliana*. Amino acids that match ECS1 are shaded in black. Note that Homologue 1 lacks an N terminal signal sequence.

FIG. 5. Alignment of the predicted amino acid sequence of *Arabidopsis* ECS1 (SEQ ID NO:2) with the predicted amino acid sequences of the four most related genes in rice and barley (SEQ ID NO:8). Amino acids that match ECS1 are shaded in black. Note that rice Homologue 2 is identical to rice Homologue 1 except it lacks the N-terminal signal sequence as seen in rice Homologue 1. The Rice ECS1, Rice ECS1/H1 and Rice ECS1/H2 sequences correspond to Rice ECS1 Homolog 2, Homolog 5 and Homolog 6, respectively (SEQ ID Nos:25, 31 and 33).

#### DETAILED DESCRIPTION OF THE INVENTION

The invention overcomes the limitations of the prior art by providing isolated nucleic acids conferring increased seed production in plants. In accordance with the invention, the nucleic acids may be introduced into selected plant species to increase seed yield. This may be achieved, for example, using developmentally-regulated promoters, or using constitutive or other desired regulatory elements.

The inventors demonstrated that heterologous overexpression of a gene designated ECS1 under a strong constitutive promoter increased the numbers of carpels and seeds per silique. Wildtype *Arabidopsis* plants have two carpels. In contrast, ECS1-overexpressing lines had three carpels, although some siliques had four carpels. Wild type plants have an average seed number of 66.2 seed/silique, whereas ECS1-overexpressing lines have 88.1 seeds per silique. The invention is therefore significant in that it may be used to increase seed production in a variety of crop species.

The ECS1 gene was first identified via its homology with a BRI1 (brassinosteroid-insensitive 1) suppressor, BRS1 (bri1 suppressor 1). BRS1 encodes a secreted serine carboxypeptidase that is implicated in an early step in brassinosteroid signaling, probably by taking part in the proteolytic processing of a protein involved in activating the BRI1 receptor (Li et al., 2001). The protease activity of BRS1 is required for its function in suppressing the phenotypes of a weak BRI1 allele, bri1-5.

BRS1 is a member of a serine carboxypeptidase gene family in *Arabidopsis*. The fact that a loss-of-function allele of BRS1 does not show any significant phenotypes suggested there is functional redundancy among the family members. To test if other members of the gene family play similar roles in suppressing the phenotypes of bri1-5, five closely related

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homologues of BRS1 were chosen and overexpressed. Three out of the five BRS1-related genes suppressed the phenotypes of bri1-5 allele. Among these three homologues, ECS1 produced an additional phenotype, i.e., increases in the numbers of carpels and seeds as described herein below.

Database searching was carried out to reveal orthologous ECS1 sequences in *Arabidopsis*, rice, barley, pea, Medicago. The sequence listing numbers of ECS1 and homologous and orthologous sequences are listed in Table 1. The five most closely related *Arabidopsis* sequences were aligned with ECS1 as shown in FIG. 4. ECS1 was 72% identical to BRS1 at the amino acid sequence level. The homologies between ECS1 and homologues 2-4 range from 52% to 60%. The homologies in the middle part of these proteins are lower than those of N-terminal and C-terminal parts. It is worth noting that homolog 1 shares 75% identity to ECS1 but lacks a N-terminal signal peptide. Interestingly, overexpression of *Arabidopsis* ECS1 homologue 1 does not suppress the bri1-5 defects and does not have the ECS1 extra-carpel silique phenotype.

The homology between ECS1, BRS1 and other type II serine carboxypeptidases indicated that ECS1 is a serine carboxypeptidase II-like protein. In addition, like BRS1, ECS1 was predicted to have an N terminal signal peptide and should be secreted. Based on its biochemical properties, yeast Kex1p is classified in the same carboxypeptidase group (carboxypeptidase D).

The regulatory roles of serine carboxypeptidases in plants have not yet been investigated. Based on an analogy with BRS1, it was predicted, without limitation to any particular mode of action, that ECS1 either process an unidentified proteinaceous proligand or a cell surface receptor (BRI1 or a BRI1 related receptor) that is involved in the control of carpel development. This processing may resemble the actions of yeast Kex1p and Kex2p, in which an *Arabidopsis* Kex2p-like endoprotease may recognize and cleave a dibasic site in its substrate. Following the cleavage, ECS1 further trims the processing intermediate, releasing either an active (co-) ligand or a functional receptor. The processing step by ECS1 may be rate limiting. Thus, elevated expression of ECS1 can increase the amount of the active form of the ligand or receptor, which subsequently enhances the signal transduction pathway involved in carpel development. As a result, extra carpels are formed and the number of seed increases.

The currently available approaches to increase seed production include traditional breeding practice (including generating hybrid plants with higher yields) and eliminating factors that reduce seed production (e.g., increasing plant's disease resistance and tolerance to various stress stimuli). It has not been shown that overexpression of the ECS1 gene, or genes that encode related carboxypeptidases, produces an increase in carpel and seed numbers in any plants.

Seed production is an essential component of crop yield. Increasing seed production has long been a pursuit of crop breeders. The invention provides a novel approach to increase seed production. After obtaining the desirable transgenic plants (i.e., plants that overexpress ECS1, its homologues or its orthologs and have been shown to have higher seed production), one can simply plant the seeds obtained from the transgenic plants without any additional manipulations. It is advantageous over traditional breeding practice, which is time-consuming and labor-intensive. Certain breeding practices require constant hybridization of desirable parent lines before seeds from hybrid plants are planted. The instant approach is also more widely applicable over those that eliminate a particular factor that reduces seed production. The transgenic plants according to the present invention may be

additionally engineered with other traits such as increased disease resistance or tolerance to cold stress that further increase their seed production. The invention may be used in agriculture to increase seed production of potentially any economically valuable plants, including, for example, soybean, *Brassica napus* (Canola/rape), rice, maize, barley, etc.

#### I. Plant Transformation Constructs

In one embodiment of the invention, plant transformation constructs are provided encoding one or more ECS1 coding sequence. By an "ECS1" sequence it is meant the nucleic acid sequences described herein capable of conferring increased seed production in plants as well as the polypeptides encoded by these sequences. Increased seed yield refers to an increase in the number of seeds and/or weight of seeds produced by a plant relative to a plant lacking a particular heterologous ECS1 coding sequence. An exemplary coding sequence for use with the invention is an *Arabidopsis thaliana* ECS1 sequence encoding the polypeptide sequence of SEQ ID NO:2. Such a coding sequence may comprise the nucleic acid sequence of SEQ ID NO:1.

Also provided by the invention are constructs encoding homologs and orthologs of the ECS1 coding sequences from both *Arabidopsis* and other plants. In certain embodiments of the invention, the orthologous sequences are from rice, barley, wheat, pea, Medicago, and *Arabidopsis*. Examples of such nucleic acids are given in SEQ ID NO:3, SEQ ID NO:5, SEQ ID NO:7; SEQ ID NO:9; SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:14, SEQ ID NO:16, SEQ ID NO:18, SEQ ID NO:20 and SEQ ID NO:22. Such nucleic acids may be further characterized as encoding a polypeptide sequence selected from the group consisting of SEQ ID NO:4, SEQ ID NO:6, SEQ ID NO:8; SEQ ID NO:10; SEQ ID NO:13, SEQ ID NO:15, SEQ ID NO:17, SEQ ID NO:19, SEQ ID NO:21 and SEQ ID NO:23.

One embodiment of the invention therefore provides a recombinant vector comprising one or more of the foregoing sequences, including all possible combinations thereof, as well as plants transformed with these sequences. Also provided by the invention are nucleic acids encoding the polypeptides encoded by these sequences, as well as polypeptides having at least about 85%, 90%, 95%, 98% and 99% amino acid identity to these sequences.

Nucleic acids that hybridize under stringent conditions to the coding sequences described herein and the use of such sequences are also provided by the invention. An example of these conditions is 5×SSC, 50% formamide and 42° C. It will be understood by those of skill in the art that stringency conditions may be increased by increasing temperature, such as to about 60° C. or decreasing salt, such as to about 1×SSC, or may be decreased by increasing salt, for example to about 10×SSC, or decreasing temperature, such as to about 25° C.

Nucleic acids provided by the invention include those encoding active ECS1 protein fragments. Those of skill in the art will immediately understand that polypeptide fragments may be prepared by placing segments of ECS1 coding sequences in frame in an appropriate expression vector, for example, comprising a plant promoter. Using the methods described in the working examples, the ability of a given polypeptide sequence to confer a phenotypic trait, such as modulation of seed production, can be efficiently confirmed for any given sequence. Fragments of nucleic acids may be prepared according to any of the well known techniques, including partial or complete restriction digests and manual shearing.

Sequences provided by the invention may be defined as encoding a functional (e.g., active) ECS1 protein. In certain

further aspects of the invention, a plant ECS1 protein may be characterized as from a monocotyledonous or dicotyledonous plant. Coding sequences may be provided operably linked to a heterologous promoter, in sense or antisense orientation. Expression constructs are also provided comprising these sequences, as are plants and plant cells transformed with the sequences.

The construction of vectors which may be employed in conjunction with plant transformation techniques using these or other sequences according to the invention will be known to those of skill of the art in light of the present disclosure (see, for example, Sambrook et al., 1989; Gelvin et al., 1990). The techniques of the current invention are thus not limited to any particular nucleic acid sequences.

One important use of the sequences provided by the invention will be in the alteration of plant phenotypes by genetic transformation with ECS1 protein coding sequences. The ECS1 protein coding sequence may be provided with other sequences for efficient expression as is known in the art. One or more selectable marker genes may be co-introduced into a plant with a nucleic acid provided by the invention.

The choice of any additional elements used in conjunction with an ECS1 coding sequence will often depend on the purpose of the transformation. One of the major purposes of transformation of crop plants is to add commercially desirable, agronomically important traits to the plant, as described above.

Vectors used for plant transformation may include, for example, plasmids, cosmids, YACs (yeast artificial chromosomes), BACs (bacterial artificial chromosomes) or any other suitable cloning system, as well as fragments of DNA therefrom. Thus when the term "vector" or "expression vector" is used, all of the foregoing types of vectors, as well as nucleic acid sequences isolated therefrom, are included. It is contemplated that utilization of cloning systems with large insert capacities will allow introduction of large DNA sequences comprising more than one selected gene. In accordance with the invention, this could be used to introduce genes corresponding to an entire biosynthetic pathway into a plant. Introduction of such sequences may be facilitated by use of bacterial or yeast artificial chromosomes (BACs or YACs, respectively), or even plant artificial chromosomes. For example, the use of BACs for *Agrobacterium*-mediated transformation was disclosed by Hamilton et al., (1996).

Particularly useful for transformation are expression cassettes which have been isolated from such vectors. DNA segments used for transforming plant cells will, of course, generally comprise the cDNA, gene or genes which one desires to introduce into and have expressed in the host cells. These DNA segments can further include structures such as promoters, enhancers, polylinkers, or regulatory genes as desired. The DNA segment or gene chosen for cellular introduction will often encode a protein which will be expressed in the resultant recombinant cells resulting in a screenable or selectable trait and/or which will impart an improved phenotype to the resulting transgenic plant. However, this may not always be the case, and the present invention also encompasses transgenic plants incorporating non-expressed transgenes. Preferred components likely to be included with vectors used in the current invention are as follows.

#### A. Regulatory Elements

Exemplary promoters for expression of a nucleic acid sequence include plant promoter such as the CaMV 35S promoter (Odell et al., 1985), or others such as CaMV 19S (Lawton et al., 1987), nos (Ebert et al., 1987), Adh (Walker et al., 1987), sucrose synthase (Yang and Russell, 1990), a-tubulin, actin (Wang et al., 1992), cab (Sullivan et al., 1989),

PEPCase (Hudspeth and Grula, 1989) or those associated with the R gene complex (Chandler et al., 1989). Tissue specific promoters such as root cell promoters (Conkling et al., 1990) and tissue specific enhancers (Fromm et al., 1986) are also contemplated to be useful, as are inducible promoters such as ABA- and turgor-inducible promoters. In one embodiment of the invention, the native promoter of an ECS1 coding sequence is used. In certain embodiments, it may be desired to employ developmentally regulated promoters such that ECS1 gene expression is triggered in concert with seed production for an increase in seed count and/or yield, but wherein expression is limited during other times.

The DNA sequence between the transcription initiation site and the start of the coding sequence, i.e., the untranslated leader sequence, can also influence gene expression. One may thus wish to employ a particular leader sequence with a transformation construct of the invention. Preferred leader sequences are contemplated to include those which comprise sequences predicted to direct optimum expression of the attached gene, i.e., to include a preferred consensus leader sequence which may increase or maintain mRNA stability and prevent inappropriate initiation of translation. The choice of such sequences will be known to those of skill in the art in light of the present disclosure. Sequences that are derived from genes that are highly expressed in plants will typically be preferred.

It is envisioned that ECS1 protein coding sequences may be introduced under the control of novel promoters or enhancers, etc., or homologous or tissue specific promoters or control elements. Vectors for use in tissue-specific targeting of genes in transgenic plants will typically include tissue-specific promoters and may also include other tissue-specific control elements such as enhancer sequences. Promoters which direct specific or enhanced expression in certain plant tissues will be known to those of skill in the art in light of the present disclosure. These include, for example, the *rbcs* promoter, specific for green tissue; the *ocs*, *nos* and *mas* promoters which have higher activity in roots or wounded leaf tissue.

#### B. Terminators

Transformation constructs prepared in accordance with the invention will typically include a 3' end DNA sequence that acts as a signal to terminate transcription and allow for the poly-adenylation of the mRNA produced by coding sequences operably linked to a promoter. In one embodiment of the invention, the native terminator of a ECS1 coding sequence is used. Alternatively, a heterologous 3' end may enhance the expression of ECS1 coding sequences. Examples of terminators that are deemed to be useful in this context include those from the nopaline synthase gene of *Agrobacterium tumefaciens* (nos 3' end) (Bevan et al., 1983), the terminator for the T7 transcript from the octopine synthase gene of *Agrobacterium tumefaciens*, and the 3' end of the protease inhibitor I or II genes from potato or tomato. Regulatory elements such as an Adh intron (Callis et al., 1987), sucrose synthase intron (Vasil et al., 1989) or TMV omega element (Gallie et al., 1989), may further be included where desired.

#### C. Transit or Signal Peptides

Sequences that are joined to the coding sequence of an expressed gene, which are removed post-translationally from the initial translation product and which facilitate the transport of the protein into or through intracellular or extracellular membranes, are termed transit (usually into vacuoles, vesicles, plastids and other intracellular organelles) and signal sequences (usually to the endoplasmic reticulum, golgi apparatus and outside of the cellular membrane). By facilitating the transport of the protein into compartments inside and outside the cell, these sequences may increase the accu-

mulation of gene product protecting them from proteolytic degradation. These sequences also allow for additional mRNA sequences from highly expressed genes to be attached to the coding sequence of the genes. Since mRNA being translated by ribosomes is more stable than naked mRNA, the presence of translatable mRNA in front of the gene may increase the overall stability of the mRNA transcript from the gene and thereby increase synthesis of the gene product. Since transit and signal sequences are usually post-translationally removed from the initial translation product, the use of these sequences allows for the addition of extra translated sequences that may not appear on the final polypeptide. It further is contemplated that targeting of certain proteins may be desirable in order to enhance the stability of the protein (U.S. Pat. No. 5,545,818, incorporated herein by reference in its entirety).

Additionally, vectors may be constructed and employed in the intracellular targeting of a specific gene product within the cells of a transgenic plant or in directing a protein to the extracellular environment. This generally will be achieved by joining a DNA sequence encoding a transit or signal peptide sequence to the coding sequence of a particular gene. The resultant transit, or signal, peptide will transport the protein to a particular intracellular, or extracellular destination, respectively, and will then be post-translationally removed.

#### D. Marker Genes

By employing a selectable marker protein, one can provide or enhance the ability to identify transformants. "Marker genes" are genes that impart a distinct phenotype to cells expressing the marker protein and thus allow such transformed cells to be distinguished from cells that do not have the marker. Such genes may encode either a selectable or screenable marker, depending on whether the marker confers a trait which one can "select" for by chemical means, i.e., through the use of a selective agent (e.g., a herbicide, antibiotic, or the like), or whether it is simply a trait that one can identify through observation or testing, i.e., by "screening" (e.g., the green fluorescent protein). Of course, many examples of suitable marker proteins are known to the art and can be employed in the practice of the invention.

Many selectable marker coding regions are known and could be used with the present invention including, but not limited to, neo (Potrykus et al., 1985), which provides kanamycin resistance and can be selected for using kanamycin, G418, paromomycin, etc.; bar, which confers bialaphos or phosphinothricin resistance; a mutant EPSP synthase protein (Hinchee et al., 1988) conferring glyphosate resistance; a nitrilase such as *bxn* from *Klebsiella ozaenae* which confers resistance to bromoxynil (Stalker et al., 1988); a mutant acetolactate synthase (ALS) which confers resistance to imidazolinone, sulfonylurea or other ALS inhibiting chemicals (European Patent Application 154, 204, 1985); a methotrexate resistant DHFR (Thillet et al., 1988), a dalapon dehalogenase that confers resistance to the herbicide dalapon; or a mutated anthranilate synthase that confers resistance to 5-methyl tryptophan.

An illustrative embodiment of selectable marker capable of being used in systems to select transformants are those that encode the enzyme phosphinothricin acetyltransferase, such as the bar gene from *Streptomyces hygroscopicus* or the pat gene from *Streptomyces viridochromogenes*. The enzyme phosphinothricin acetyl transferase (PAT) inactivates the active ingredient in the herbicide bialaphos, phosphinothricin (PPT). PPT inhibits glutamine synthetase, (Murakami et al., 1986; Twell et al., 1989) causing rapid accumulation of ammonia and cell death.

Screenable markers that may be employed include a  $\beta$ -glucuronidase (GUS) or uidA gene which encodes an enzyme for which various chromogenic substrates are known; an R-locus gene, which encodes a product that regulates the production of anthocyanin pigments (red color) in plant tissues (Delaporta et al., 1988); a  $\beta$ -lactamase gene (Sutcliffe, 1978), which encodes an enzyme for which various chromogenic substrates are known (e.g., PADAC, a chromogenic cephalosporin); a xyle gene (Zukowsky et al., 1983) which encodes a catechol dioxygenase that can convert chromogenic catechols; an  $\alpha$ -amylase gene (Ikuta et al., 1990); a tyrosinase gene (Katz et al., 1983) which encodes an enzyme capable of oxidizing tyrosine to DOPA and dopaquinone which in turn condenses to form the easily-detectable compound melanin; a  $\beta$ -galactosidase gene, which encodes an enzyme for which there are chromogenic substrates; a luciferase (lux) gene (Ow et al., 1986), which allows for bioluminescence detection; an aequorin gene (Prasher et al., 1985) which may be employed in calcium-sensitive bioluminescence detection; or a gene encoding for green fluorescent protein (Sheen et al., 1995; Haseloff et al., 1997; Reichel et al., 1996; Tian et al., 1997; WO 97/41228). The gene that encodes green fluorescent protein (GFP) is also contemplated as a particularly useful reporter gene (Sheen et al., 1995; Haseloff et al., 1997; Reichel et al., 1996; Tian et al., 1997; WO 97/41228). Expression of green fluorescent protein may be visualized in a cell or plant as fluorescence following illumination by particular wavelengths of light.

## II. Methods for Genetic Transformation

Suitable methods for transformation of plant or other cells for use with the current invention are believed to include virtually any method by which DNA can be introduced into a cell, such as by direct delivery of DNA such as by PEG-mediated transformation of protoplasts (Omirulleh et al., 1993), by desiccation/inhibition-mediated DNA uptake (Potrykus et al., 1985), by electroporation (U.S. Pat. No. 5,384,253, specifically incorporated herein by reference in its entirety), by agitation with silicon carbide fibers (Kaeppeler et al., 1990; U.S. Pat. No. 5,302,523, specifically incorporated herein by reference in its entirety; and U.S. Pat. No. 5,464,765, specifically incorporated herein by reference in its entirety), by *Agrobacterium*-mediated transformation (U.S. Pat. No. 5,591,616 and U.S. Pat. No. 5,563,055; both specifically incorporated herein by reference) and by acceleration of DNA coated particles (U.S. Pat. No. 5,550,318; U.S. Pat. No. 5,538,877; and U.S. Pat. No. 5,538,880; each specifically incorporated herein by reference in its entirety), etc. Through the application of techniques such as these, the cells of virtually any plant species may be stably transformed, and these cells developed into transgenic plants.

A. *Agrobacterium*-mediated Transformation *Agrobacterium*-mediated transfer is a widely applicable system for introducing genes into plant cells because the DNA can be introduced into whole plant tissues, thereby bypassing the need for regeneration of an intact plant from a protoplast. The use of *Agrobacterium*-mediated plant integrating vectors to introduce DNA into plant cells is well known in the art. See, for example, the methods described by Fraley et al., (1985), Rogers et al., (1987) and U.S. Pat. No. 5,563,055, specifically incorporated herein by reference in its entirety.

*Agrobacterium*-mediated transformation is most efficient in dicotyledonous plants and is the preferable method for transformation of dicots, including *Arabidopsis*, tobacco, tomato, alfalfa and potato. Indeed, while *Agrobacterium*-mediated transformation has been routinely used with dicotyledonous plants for a number of years, it has only recently

become applicable to monocotyledonous plants. Advances in *Agrobacterium*-mediated transformation techniques have now made the technique applicable to nearly all monocotyledonous plants. For example, *Agrobacterium*-mediated transformation techniques have now been applied to rice (Hiei et al., 1997; U.S. Pat. No. 5,591,616, specifically incorporated herein by reference in its entirety), wheat (McCormac et al., 1998), barley (Tingay et al., 1997; McCormac et al., 1998), alfalfa (Thomas et al., 1990) and maize (Ishidia et al., 1996).

Modern *Agrobacterium* transformation vectors are capable of replication in *E. coli* as well as *Agrobacterium*, allowing for convenient manipulations as described (Klee et al., 1985). Moreover, recent technological advances in vectors for *Agrobacterium*-mediated gene transfer have improved the arrangement of genes and restriction sites in the vectors to facilitate the construction of vectors capable of expressing various polypeptide coding genes. The vectors described (Rogers et al., 1987) have convenient multi-linker regions flanked by a promoter and a polyadenylation site for direct expression of inserted polypeptide coding genes and are suitable for present purposes. In addition, *Agrobacterium* containing both armed and disarmed Ti genes can be used for the transformations. In those plant strains where *Agrobacterium*-mediated transformation is efficient, it is the method of choice because of the facile and defined nature of the gene transfer.

### B. Electroporation

To effect transformation by electroporation, one may employ either friable tissues, such as a suspension culture of cells or embryogenic callus or alternatively one may transform immature embryos or other organized tissue directly. In this technique, one would partially degrade the cell walls of the chosen cells by exposing them to pectin-degrading enzymes (pectolyases) or mechanically wounding in a controlled manner. Examples of some species which have been transformed by electroporation of intact cells include maize (U.S. Pat. No. 5,384,253; Rhodes et al., 1995; D'Halluin et al., 1992), wheat (Zhou et al., 1993), tomato (Hou and Lin, 1996), soybean (Christou et al., 1987) and tobacco (Lee et al., 1989).

One also may employ protoplasts for electroporation transformation of plants (Bates, 1994; Lazzeri, 1995). For example, the generation of transgenic soybean plants by electroporation of cotyledon-derived protoplasts is described by Dhir and Widholm in Intl. Patent Appl. Publ. No. WO 92/17598 (specifically incorporated herein by reference). Other examples of species for which protoplast transformation has been described include barley (Lazzeri, 1995), sorghum (Battraw et al., 1991), maize (Bhattacharjee et al., 1997), wheat (He et al., 1994) and tomato (Tsukada, 1989).

### C. Microprojectile Bombardment

Another method for delivering transforming DNA segments to plant cells in accordance with the invention is microprojectile bombardment (U.S. Pat. No. 5,550,318; U.S. Pat. No. 5,538,880; U.S. Pat. No. 5,610,042; and PCT Application WO 94/09699; each of which is specifically incorporated herein by reference in its entirety). In this method, particles may be coated with nucleic acids and delivered into cells by a propelling force. Exemplary particles include those comprised of tungsten, platinum, and preferably, gold. It is contemplated that in some instances DNA precipitation onto metal particles would not be necessary for DNA delivery to a recipient cell using microprojectile bombardment. However, it is contemplated that particles may contain DNA rather than be coated with DNA. Hence, it is proposed that DNA-coated particles may increase the level of DNA delivery via particle bombardment but are not, in and of themselves, necessary.

For the bombardment, cells in suspension are concentrated on filters or solid culture medium. Alternatively, immature embryos or other target cells may be arranged on solid culture medium. The cells to be bombarded are positioned at an appropriate distance below the macroprojectile stopping plate.

An illustrative embodiment of a method for delivering DNA into plant cells by acceleration is the Biolistics Particle Delivery System, which can be used to propel particles coated with DNA or cells through a screen, such as a stainless steel or Nytex screen, onto a filter surface covered with monocot plant cells cultured in suspension. The screen disperses the particles so that they are not delivered to the recipient cells in large aggregates. Microprojectile bombardment techniques are widely applicable, and may be used to transform virtually any plant species. Examples of species for which have been transformed by microprojectile bombardment include monocot species such as maize (PCT Application WO 95/06128), barley (Ritala et al., 1994; Hensgens et al., 1993), wheat (U.S. Pat. No. 5,563,055, specifically incorporated herein by reference in its entirety), rice (Hensgens et al., 1993), oat (Torbet et al., 1995; Torbet et al., 1998), rye (Hensgens et al., 1993), sugarcane (Bower et al., 1992), and sorghum (Casa et al., 1993; Hagio et al., 1991); as well as a number of dicots including tobacco (Tomes et al., 1990; Buising and Benbow, 1994), soybean (U.S. Pat. No. 5,322,783, specifically incorporated herein by reference in its entirety), sunflower (Knittel et al., 1994), peanut (Singsit et al., 1997), cotton (McCabe and Martinell, 1993), tomato (VanEck et al., 1995), and legumes in general (U.S. Pat. No. 5,563,055, specifically incorporated herein by reference in its entirety).

#### D. Other Transformation Methods

Transformation of protoplasts can be achieved using methods based on calcium phosphate precipitation, polyethylene glycol treatment, electroporation, and combinations of these treatments (see, e.g., Potrykus et al., 1985; Lorz et al., 1985; Omirulleh et al., 1993; Fromm et al., 1986; Uchimiya et al., 1986; Callis et al., 1987; Marcotte et al., 1988).

Application of these systems to different plant strains depends upon the ability to regenerate that particular plant strain from protoplasts. Illustrative methods for the regeneration of cereals from protoplasts have been described (Toriyama et al., 1986; Yamada et al., 1986; Abdullah et al., 1986; Omirulleh et al., 1993 and U.S. Pat. No. 5,508,184; each specifically incorporated herein by reference in its entirety). Examples of the use of direct uptake transformation of cereal protoplasts include transformation of rice (Ghosh-Biswas et al., 1994), sorghum (Battraw and Hall, 1991), barley (Lazerri, 1995), oat (Zheng and Edwards, 1990) and maize (Omirulleh et al., 1993).

To transform plant strains that cannot be successfully regenerated from protoplasts, other ways to introduce DNA into intact cells or tissues can be utilized. For example, regeneration of cereals from immature embryos or explants can be effected as described (Vasil, 1989). Also, silicon carbide fiber-mediated transformation may be used with or without protoplasting (Kaeppeler, 1990; Kaeppeler et al., 1992; U.S. Pat. No. 5,563,055, specifically incorporated herein by reference in its entirety). Transformation with this technique is accomplished by agitating silicon carbide fibers together with cells in a DNA solution. DNA passively enters as the cells are punctured. This technique has been used successfully with, for example, the monocot cereals maize (PCT Application WO 95/06128, specifically incorporated herein by reference in its entirety; (Thompson, 1995) and rice (Nagatani, 1997).

#### E. Tissue Cultures

Tissue cultures may be used in certain transformation techniques for the preparation of cells for transformation and for the regeneration of plants therefrom. Maintenance of tissue cultures requires use of media and controlled environments. "Media" refers to the numerous nutrient mixtures that are used to grow cells in vitro, that is, outside of the intact living organism. The medium usually is a suspension of various categories of ingredients (salts, amino acids, growth regulators, sugars, buffers) that are required for growth of most cell types. However, each specific cell type requires a specific range of ingredient proportions for growth, and an even more specific range of formulas for optimum growth. Rate of cell growth also will vary among cultures initiated with the array of media that permit growth of that cell type.

Nutrient media is prepared as a liquid, but this may be solidified by adding the liquid to materials capable of providing a solid support. Agar is most commonly used for this purpose. Bactoagar, Hazelton agar, Gelrite, and Gelgro are specific types of solid support that are suitable for growth of plant cells in tissue culture.

Some cell types will grow and divide either in liquid suspension or on solid media. As disclosed herein, plant cells will grow in suspension or on solid medium, but regeneration of plants from suspension cultures typically requires transfer from liquid to solid media at some point in development. The type and extent of differentiation of cells in culture will be affected not only by the type of media used and by the environment, for example, pH, but also by whether media is solid or liquid.

Somatic cells are of various types. Embryogenic cells are one example of somatic cells which may be induced to regenerate a plant through embryo formation. Non-embryogenic cells are those which typically will not respond in such a fashion. Certain techniques may be used that enrich recipient cells within a cell population. For example, Type II callus development, followed by manual selection and culture of friable, embryogenic tissue, generally results in an enrichment of cells. Manual selection techniques which can be employed to select target cells may include, e.g., assessing cell morphology and differentiation, or may use various physical or biological means. Cryopreservation also is a possible method of selecting for recipient cells.

Where employed, cultured cells may be grown either on solid supports or in the form of liquid suspensions. In either instance, nutrients may be provided to the cells in the form of media, and environmental conditions controlled. There are many types of tissue culture media comprised of various amino acids, salts, sugars, growth regulators and vitamins. Most of the media employed in the practice of the invention will have some similar components, but may differ in the composition and proportions of their ingredients depending on the particular application envisioned. For example, various cell types usually grow in more than one type of media, but will exhibit different growth rates and different morphologies, depending on the growth media. In some media, cells survive but do not divide. Various types of media suitable for culture of plant cells previously have been described. Examples of these media include, but are not limited to, the N6 medium described by Chu et al., (1975) and MS media (Murashige and Skoog, 1962).

#### III. Production and Characterization of Stably Transformed Plants

After effecting delivery of exogenous DNA to recipient cells, the next steps generally concern identifying the transformed cells for further culturing and plant regeneration. In

order to improve the ability to identify transformants, one may desire to employ a selectable or screenable marker gene with a transformation vector prepared in accordance with the invention. In this case, one would then generally assay the potentially transformed cell population by exposing the cells to a selective agent or agents, or one would screen the cells for the desired marker gene trait.

#### A. Selection

It is believed that DNA is introduced into only a small percentage of target cells in any one study. In order to provide an efficient system for identification of those cells receiving DNA and integrating it into their genomes one may employ a means for selecting those cells that are stably transformed. One exemplary embodiment of such a method is to introduce into the host cell, a marker gene which confers resistance to some normally inhibitory agent, such as an antibiotic or herbicide. Examples of antibiotics which may be used include the aminoglycoside antibiotics neomycin, kanamycin and paromomycin, or the antibiotic hygromycin. Resistance to the aminoglycoside antibiotics is conferred by aminoglycoside phosphotransferase enzymes such as neomycin phosphotransferase II (NPT II) or NPT I, whereas resistance to hygromycin is conferred by hygromycin phosphotransferase.

Potentially transformed cells then are exposed to the selective agent. In the population of surviving cells will be those cells where, generally, the resistance-conferring gene has been integrated and expressed at sufficient levels to permit cell survival. Cells may be tested further to confirm stable integration of the exogenous DNA.

One herbicide which constitutes a desirable selection agent is the broad spectrum herbicide bialaphos. Bialaphos is a tripeptide antibiotic produced by *Streptomyces hygroscopicus* and is composed of phosphinothricin (PPT), an analogue of L-glutamic acid, and two L-alanine residues. Upon removal of the L-alanine residues by intracellular peptidases, the PPT is released and is a potent inhibitor of glutamine synthetase (GS), a pivotal enzyme involved in ammonia assimilation and nitrogen metabolism (Ogawa et al., 1973). Synthetic PPT, the active ingredient in the herbicide Liberty™ also is effective as a selection agent. Inhibition of GS in plants by PPT causes the rapid accumulation of ammonia and death of the plant cells.

The organism producing bialaphos and other species of the genus *Streptomyces* also synthesizes an enzyme phosphinothricin acetyl transferase (PAT) which is encoded by the bar gene in *Streptomyces hygroscopicus* and the pat gene in *Streptomyces viridochromogenes*. The use of the herbicide resistance gene encoding phosphinothricin acetyl transferase (PAT) is referred to in DE 3642 829 A, wherein the gene is isolated from *Streptomyces viridochromogenes*. In the bacterial source organism, this enzyme acetylates the free amino group of PPT preventing auto-toxicity (Thompson et al., 1987). The bar gene has been cloned (Murakami et al., 1986; Thompson et al., 1987) and expressed in transgenic tobacco, tomato, potato (De Block et al., 1987) *Brassica* (De Block et al., 1989) and maize (U.S. Pat. No. 5,550,318). In previous reports, some transgenic plants which expressed the resistance gene were completely resistant to commercial formulations of PPT and bialaphos in greenhouses.

Another example of a herbicide which is useful for selection of transformed cell lines in the practice of the invention is the broad spectrum herbicide glyphosate. Glyphosate inhibits the action of the enzyme EPSPS which is active in the aromatic amino acid biosynthetic pathway. Inhibition of this enzyme leads to starvation for the amino acids phenylalanine, tyrosine, and tryptophan and secondary metabolites derived thereof. U.S. Pat. No. 4,535,060 describes the isolation of

EPSPS mutations which confer glyphosate resistance on the *Salmonella typhimurium* gene for EPSPS, aroA. The EPSPS gene was cloned from *Zea mays* and mutations similar to those found in a glyphosate resistant aroA gene were introduced in vitro. Mutant genes encoding glyphosate resistant EPSPS enzymes are described in, for example, International Patent WO 97/4103. The best characterized mutant EPSPS gene conferring glyphosate resistance comprises amino acid changes at residues 102 and 106, although it is anticipated that other mutations will also be useful (PCT/WO97/4103).

To use the bar-bialaphos or the EPSPS-glyphosate selective system, transformed tissue is cultured for 0-28 days on nonselective medium and subsequently transferred to medium containing from 1-3 mg/l bialaphos or 1-3 mM glyphosate as appropriate. While ranges of 1-3 mg/l bialaphos or 1-3 mM glyphosate will typically be preferred, it is proposed that ranges of 0.1-50 mg/l bialaphos or 0.1-50 mM glyphosate will find utility.

An example of a screenable marker trait is the enzyme luciferase. In the presence of the substrate luciferin, cells expressing luciferase emit light which can be detected on photographic or x-ray film, in a luminometer (or liquid scintillation counter), by devices that enhance night vision, or by a highly light sensitive video camera, such as a photon counting camera. These assays are nondestructive and transformed cells may be cultured further following identification. The photon counting camera is especially valuable as it allows one to identify specific cells or groups of cells which are expressing luciferase and manipulate those in real time. Another screenable marker which may be used in a similar fashion is the gene coding for green fluorescent protein.

#### B. Regeneration and Seed Production

Cells that survive the exposure to the selective agent, or cells that have been scored positive in a screening assay, may be cultured in media that supports regeneration of plants. In an exemplary embodiment, MS and N6 media may be modified by including further substances such as growth regulators. One such growth regulator is dicamba or 2,4-D. However, other growth regulators may be employed, including NAA, NAA+2,4-D or picloram. Media improvement in these and like ways has been found to facilitate the growth of cells at specific developmental stages. Tissue may be maintained on a basic media with growth regulators until sufficient tissue is available to begin plant regeneration efforts, or following repeated rounds of manual selection, until the morphology of the tissue is suitable for regeneration, at least 2 wk, then transferred to media conducive to maturation of embryoids. Cultures are transferred every 2 wk on this medium. Shoot development will signal the time to transfer to medium lacking growth regulators.

The transformed cells, identified by selection or screening and cultured in an appropriate medium that supports regeneration, will then be allowed to mature into plants. Developing plantlets are transferred to soilless plant growth mix, and hardened, e.g., in an environmentally controlled chamber, for example, at about 85% relative humidity, 600 ppm CO<sub>2</sub>, and 25-250 microeinsteins m<sup>-2</sup> s<sup>-1</sup> of light. Plants may be matured in a growth chamber or greenhouse. Plants can be regenerated from about 6 wk to 10 months after a transformant is identified, depending on the initial tissue. During regeneration, cells are grown on solid media in tissue culture vessels. Illustrative embodiments of such vessels are petri dishes and Plant Cons. Regenerating plants can be grown at about 19 to 28° C. After the regenerating plants have reached the stage of shoot and root development, they may be transferred to a greenhouse for further growth and testing.

Seeds on transformed plants may occasionally require embryo rescue due to cessation of seed development and premature senescence of plants. To rescue developing embryos, they are excised from surface-disinfected seeds 10-20 days post-pollination and cultured. An embodiment of media used for culture at this stage comprises MS salts, 2% sucrose, and 5.5 g/l agarose. In embryo rescue, large embryos (defined as greater than 3 mm in length) are germinated directly on an appropriate media. Embryos smaller than that may be cultured for 1 wk on media containing the above ingredients along with  $10^{-5}$ M abscisic acid and then transferred to growth regulator-free medium for germination.

#### C. Characterization

To confirm the presence of the exogenous DNA or "trans-gene(s)" in the regenerating plants, a variety of assays may be performed. Such assays include, for example, "molecular biological" assays, such as Southern and Northern blotting and PCR<sup>TM</sup>; "biochemical" assays, such as detecting the presence of a protein product, e.g., by immunological means (ELISAs and Western blots) or by enzymatic function; plant part assays, such as leaf or root assays; and also, by analyzing the phenotype of the whole regenerated plant.

#### D. DNA Integration, RNA Expression and Inheritance

Genomic DNA may be isolated from cell lines or any plant parts to determine the presence of the exogenous gene through the use of techniques well known to those skilled in the art. Note, that intact sequences will not always be present, presumably due to rearrangement or deletion of sequences in the cell. The presence of DNA elements introduced through the methods of this invention may be determined, for example, by polymerase chain reaction (PCR<sup>TM</sup>). Using this technique, discreet fragments of DNA are amplified and detected by gel electrophoresis. This type of analysis permits one to determine whether a gene is present in a stable transformant, but does not prove integration of the introduced gene into the host cell genome. It is typically the case, however, that DNA has been integrated into the genome of all transformants that demonstrate the presence of the gene through PCR<sup>TM</sup> analysis. In addition, it is not typically possible using PCR<sup>TM</sup> techniques to determine whether transformants have exogenous genes introduced into different sites in the genome, i.e., whether transformants are of independent origin. It is contemplated that using PCR<sup>TM</sup> techniques it would be possible to clone fragments of the host genomic DNA adjacent to an introduced gene.

Positive proof of DNA integration into the host genome and the independent identities of transformants may be determined using the technique of Southern hybridization. Using this technique specific DNA sequences that were introduced into the host genome and flanking host DNA sequences can be identified. Hence the Southern hybridization pattern of a given transformant serves as an identifying characteristic of that transformant. In addition it is possible through Southern hybridization to demonstrate the presence of introduced genes in high molecular weight DNA, i.e., confirm that the introduced gene has been integrated into the host cell genome. The technique of Southern hybridization provides information that is obtained using PCR<sup>TM</sup>, e.g., the presence of a gene, but also demonstrates integration into the genome and characterizes each individual transformant.

It is contemplated that using the techniques of dot or slot blot hybridization which are modifications of Southern hybridization techniques one could obtain the same information that is derived from PCR<sup>TM</sup>, e.g., the presence of a gene.

Both PCR<sup>TM</sup> and Southern hybridization techniques can be used to demonstrate transmission of a transgene to progeny.

In most instances the characteristic Southern hybridization pattern for a given transformant will segregate in progeny as one or more Mendelian genes (Spencer et al., 1992) indicating stable inheritance of the transgene.

Whereas DNA analysis techniques may be conducted using DNA isolated from any part of a plant, RNA will only be expressed in particular cells or tissue types and hence it will be necessary to prepare RNA for analysis from these tissues. PCR<sup>TM</sup> techniques also may be used for detection and quantitation of RNA produced from introduced genes. In this application of PCR<sup>TM</sup> it is first necessary to reverse transcribe RNA into DNA, using enzymes such as reverse transcriptase, and then through the use of conventional PCR<sup>TM</sup> techniques amplify the DNA. In most instances PCR<sup>TM</sup> techniques, while useful, will not demonstrate integrity of the RNA product. Further information about the nature of the RNA product may be obtained by Northern blotting. This technique will demonstrate the presence of an RNA species and give information about the integrity of that RNA. The presence or absence of an RNA species also can be determined using dot or slot blot Northern hybridizations. These techniques are modifications of Northern blotting and will only demonstrate the presence or absence of an RNA species.

#### E. Gene Expression

While Southern blotting and PCR<sup>TM</sup> may be used to detect the gene(s) in question, they do not provide information as to whether the corresponding protein is being expressed. Expression may be evaluated by specifically identifying the protein products of the introduced genes or evaluating the phenotypic changes brought about by their expression.

Assays for the production and identification of specific proteins may make use of physical-chemical, structural, functional, or other properties of the proteins. Unique physical-chemical or structural properties allow the proteins to be separated and identified by electrophoretic procedures, such as native or denaturing gel electrophoresis or isoelectric focusing, or by chromatographic techniques such as ion exchange or gel exclusion chromatography. The unique structures of individual proteins offer opportunities for use of specific antibodies to detect their presence in formats such as an ELISA assay. Combinations of approaches may be employed with even greater specificity such as western blotting in which antibodies are used to locate individual gene products that have been separated by electrophoretic techniques. Additional techniques may be employed to absolutely confirm the identity of the product of interest such as evaluation by amino acid sequencing following purification. Although these are among the most commonly employed, other procedures may be additionally used.

Assay procedures also may be used to identify the expression of proteins by their functionality, especially the ability of enzymes to catalyze specific chemical reactions involving specific substrates and products. These reactions may be followed by providing and quantifying the loss of substrates or the generation of products of the reactions by physical or chemical procedures. Examples are as varied as the enzyme to be analyzed and may include assays for PAT enzymatic activity by following production of radiolabeled acetylated phosphinothricin from phosphinothricin and <sup>14</sup>C-acetyl CoA or for anthranilate synthase activity by following loss of fluorescence of anthranilate, to name two.

Very frequently the expression of a gene product is determined by evaluating the phenotypic results of its expression. These assays also may take many forms including but not limited to analyzing changes in the chemical composition, morphology, or physiological properties of the plant. Chemical composition may be altered by expression of genes encod-



ing enzymes or storage proteins which change amino acid composition and may be detected by amino acid analysis, or by enzymes which change starch quantity which may be analyzed by near infrared reflectance spectrometry. Morphological changes may include greater stature or thicker stalks. Most often changes in response of plants or plant parts to imposed treatments are evaluated under carefully controlled conditions termed bioassays.

#### IV. Breeding Plants of the Invention

In addition to direct transformation of a particular plant genotype with a construct prepared according to the current invention, transgenic plants may be made by crossing a plant having a selected DNA of the invention to a second plant lacking the construct. For example, a selected coding sequence can be introduced into a particular plant variety by crossing, without the need for ever directly transforming a plant of that given variety. Therefore, the current invention not only encompasses a plant directly transformed or regenerated from cells which have been transformed in accordance with the current invention, but also the progeny of such plants.

As used herein the term "progeny" denotes the offspring of any generation of a parent plant prepared in accordance with the instant invention, wherein the progeny comprises a selected DNA construct. "Crossing" a plant to provide a plant line having one or more added transgenes relative to a starting plant line, as disclosed herein, is defined as the techniques that result in a transgene of the invention being introduced into a plant line by crossing a starting line with a donor plant line that comprises a transgene of the invention. To achieve this one could, for example, perform the following steps:

- (a) plant seeds of the first (starting line) and second (donor plant line that comprises a transgene of the invention) parent plants;
- (b) grow the seeds of the first and second parent plants into plants that bear flowers;
- (c) pollinate a flower from the first parent plant with pollen from the second parent plant; and
- (d) harvest seeds produced on the parent plant bearing the fertilized flower.

Backcrossing is herein defined as the process including the steps of:

- (a) crossing a plant of a first genotype containing a desired gene, DNA sequence or element to a plant of a second genotype lacking the desired gene, DNA sequence or element;
- (b) selecting one or more progeny plant containing the desired gene, DNA sequence or element;
- (c) crossing the progeny plant to a plant of the second genotype; and
- (d) repeating steps (b) and (c) for the purpose of transferring a desired DNA sequence from a plant of a first genotype to a plant of a second genotype.

Introgression of a DNA element into a plant genotype is defined as the result of the process of backcross conversion. A plant genotype into which a DNA sequence has been introgressed may be referred to as a backcross converted genotype, line, inbred, or hybrid. Similarly a plant genotype lacking the desired DNA sequence may be referred to as an unconverted genotype, line, inbred, or hybrid.

#### V. Definitions

**Expression:** The combination of intracellular processes, including transcription and translation undergone by a coding DNA molecule such as a structural gene to produce a polypeptide.

**Genetic Transformation:** A process of introducing a DNA sequence or construct (e.g., a vector or expression cassette)

into a cell or protoplast in which that exogenous DNA is incorporated into a chromosome or is capable of autonomous replication.

**Heterologous:** A sequence which is not normally present in a given host genome in the genetic context in which the sequence is currently found. In this respect, the sequence may be native to the host genome, but be rearranged with respect to other genetic sequences within the host sequence. For example, a coding sequence may be heterologous in that it is linked to a different promoter sequence relative to the native coding sequence.

**Obtaining:** When used in conjunction with a transgenic plant cell or transgenic plant, obtaining means either transforming a non-transgenic plant cell or plant to create the transgenic plant cell or plant, or planting transgenic plant seed to produce the transgenic plant cell or plant. Such a transgenic plant seed may be from an  $R_0$  transgenic plant or may be from a progeny of any generation thereof that inherits a given transgenic sequence from a starting transgenic parent plant.

**Promoter:** A recognition site on a DNA sequence or group of DNA sequences that provides an expression control element for a structural gene and to which RNA polymerase specifically binds and initiates RNA synthesis (transcription) of that gene.

**$R_0$  transgenic plant:** A plant that has been genetically transformed or has been regenerated from a plant cell or cells that have been genetically transformed.

**Regeneration:** The process of growing a plant from a plant cell (e.g., plant protoplast, callus or explant).

**Selected DNA:** A DNA segment which one desires to introduce or has introduced into a plant genome by genetic transformation.

**Transformation construct:** A chimeric DNA molecule which is designed for introduction into a host genome by genetic transformation. Preferred transformation constructs will comprise all of the genetic elements necessary to direct the expression of one or more exogenous genes. In particular embodiments of the instant invention, it may be desirable to introduce a transformation construct into a host cell in the form of an expression cassette.

**Transformed cell:** A cell the DNA complement of which has been altered by the introduction of an exogenous DNA molecule into that cell.

**Transgene:** A segment of DNA which has been incorporated into a host genome or is capable of autonomous replication in a host cell and is capable of causing the expression of one or more coding sequences. Exemplary transgenes will provide the host cell, or plants regenerated therefrom, with a novel phenotype relative to the corresponding non-transformed cell or plant. Transgenes may be directly introduced into a plant by genetic transformation, or may be inherited from a plant of any previous generation which was transformed with the DNA segment.

**Transgenic plant:** A plant or progeny plant of any subsequent generation derived therefrom, wherein the DNA of the plant or progeny thereof contains an introduced exogenous DNA segment not naturally present in a non-transgenic plant of the same strain. The transgenic plant may additionally contain sequences which are native to the plant being transformed, but wherein the "exogenous" gene has been altered in order to alter the level or pattern of expression of the gene, for example, by use of one or more heterologous regulatory or other elements.

**Vector:** A DNA molecule designed for transformation into a host cell. Some vectors may be capable of replication in a host cell. A plasmid is an exemplary vector, as are expression cassettes isolated therefrom.



VI. Examples

The following examples are included to demonstrate preferred embodiments of the invention. It should be appreciated by those of skill in the art that the techniques disclosed in the examples which follow represent techniques discovered by the inventors to function well in the practice of the invention, and thus can be considered to constitute preferred modes for its practice. However, those of skill in the art should, in light of the present disclosure, appreciate that many changes can be made in the specific embodiments which are disclosed and still obtain a like or similar result without departing from the concept, spirit and scope of the invention. More specifically, it will be apparent that certain agents which are both chemically and physiologically related may be substituted for the agents described herein while the same or similar results would be achieved. All such similar substitutes and modifications apparent to those skilled in the art are deemed to be within the spirit, scope and concept of the invention as defined by the appended claims.

EXAMPLE 1

Identification of ECS1, a Gene Conferring Increased Seed Production

BRS1 encodes a secreted serine carboxypeptidase that is implicated in an early step in brassinosteroid signaling, and is indicated as taking part in the proteolytic processing of a protein involved in activating the BRI1 receptor (Li et al., 2001). The protease activity of BRS1 is required for its function in suppressing the phenotypes of a weak BRI1 allele, bri1-5. BRS1 is a member of a serine carboxypeptidase gene family in *Arabidopsis*. The fact that a loss-of-function allele of BRS1 does not show any significant phenotypes suggested there is functional redundancy among the family members.

To test if other members of the gene family play similar roles in suppressing the phenotypes of bri1-5, five closely related homologues of BRS1 were chosen and the corresponding cDNAs of these homologues expressed under a 35S promoter in bri1-5 plants by *Agrobacterium*-mediated transformation (Clough and Bent 1998). Results showed that three out of the five BRS1-related genes suppressed the phenotype of the bri1-5 allele. Among these three homologues, ECS1 produced an additional phenotype, i.e., increases in the numbers of carpels and seeds as described in more detail below.

EXAMPLE 2

Overexpression of ECS1

Overexpressing ECS1 under a strong constitutive promoter in wild type *Arabidopsis* plants was demonstrated to increase the numbers of carpels and seeds per silique (FIG. 1). Wild-type *Arabidopsis* plants have two carpels. In contrast, ECS1-overexpressing lines had three carpels, although some siliques had four carpels. Wild type plants have an average seed number of 66.2 seed/silique, whereas ECS1-overexpressing lines had 88.1 seeds per silique. The weight of 1000 seeds from ECS1-overexpressing plants was not significantly different from that of wildtype, showing that these seeds are of normal size and shape. However, the total seed weight/silique was increased by about 33% in ECS1-overexpressing plants due to the increased total number of seeds.

As can be seen in FIG. 1, wildtype plants had a low level of ECS1, while the ECS1-overexpressing line (in WS background) had an elevated ECS1 expression level (top panel). ACT7 was used as a probe to show the sample equal loading of total RNA (bottom panel).

The overexpression of ECS1 suppressed the bri1-5 phenotype. Rosette leaves in bri1-5 are curled, while ECS1-overexpressing plants have expanded leaves. bri1-5 plants flower 7-10 days later than wildtype plants, while ECS1-overexpressing plants flower 5-7 days earlier than bri1-5 plant. Interestingly, ECS1-overexpressing lines in bri1-5 had four-carpel siliques (FIG. 3). Carpels are the ovule (seed)-bearing organ in gynoecium, and the increased carpel numbers lead to elevated seed numbers per silique. A two-carpel silique from bri1-5 plants has an average of 43.2 seeds, while the four-carpel silique from ECS1-overexpressing plants increased the seed number to 58.3 seeds/silique.

Data was collected regarding seed yield in a population of ECS1-overexpressing transgenic plants (35S::ECS1; 28 plants) and a population of wild-type plants (29 plants) grown to maturity in the greenhouse. Total seed was collected from each individual and weighed to determine total seed yield per plant (Table 1).

TABLE 1

analysis of total seed yield.		
	35S::ECS1	Wild-type
Mean seed weight/plant (gm)	.82	.77
SD (gm)	.16	.13
N	28	29

P value = .178

Statistical analysis of the data indicated that the seed yield from the two populations was not statistically different. Because the 35S::ECS1 plants produce more seed per fruit, this result suggests that the ECS1-overexpressing plants have fewer fruit per plant. This would be consistent with qualitative observations that the 35S::ECS1 plants used in this study were somewhat smaller than wildtype and appeared to produce fewer flowers.

There were several possible explanations for why an increase in total seed yield per plant was not observed. The 35S::ECS1 transgenic lines used in this study were all siblings and the result may be due to a transgene position effect. Several independent 35S::ECS1 lines were analyzed in the bri1-5 background and the increased carpel number and seed per fruit was consistent. In addition, there were likely background differences between the 35S::ECS1 transgenic lines and the wild type. The 35S::ECS1 transgenic line is the result of crossing 35S::ECS1 bri1-5 with wild type and isolating plants that were wildtype for BRI1. To control for these variables, additional, independent 35S::ECS1 lines are being generated in the Col ecotype for comparison of total seed yield between these lines and the Col wildtype. The use of tissues specific promoters to limit ECS1 expression in flowers and fruits will also be analyzed.

EXAMPLE 3

Identification of Orthologous Plant Coding Sequences

Database searching was carried out to reveal ECS1 sequences in *Arabidopsis*, rice, barley, pea, Medicago. The sequence database accession numbers of ECS1 and some of its homologs and orthologs identified are listed in Table 1. The five most closely related *Arabidopsis* sequences are aligned with ECS1 in FIG. 4. Amino acids that match ECS1 are shaded in black. ECS1 is 72% identical to BRS1 at the amino acid sequence level. The homologies between ECS1

and homologs 2-4 range from 52% to 60%. The homologies in the middle part of these proteins are lower than those of N-terminal and C-terminal parts. It is worth noting that homolog 1 shares 75% identity to ECS1 but lacks a N-terminal signal peptide. Interestingly, overexpression of *Arabidopsis* ECS1 homologue 1 does not suppress the *bri1-5* defects and does not have the ECS1 silique phenotype.

TABLE 2

Sequence Database Accession Numbers of <i>Arabidopsis</i> ECS1 and its Homologues and Orthologs		
Name	Accession Number	SEQ ID NO
<i>Arabidopsis</i> ECS1	AAC63668.1	SEQ ID NOS: 1-2
<i>Arabidopsis</i> ECS1 homolog 1	AAC63669.1	SEQ ID NO: 16
<i>Arabidopsis</i> ECS1 homolog 2	AAO11573.1 and AAM65698.1	SEQ ID NO: 17
<i>Arabidopsis</i> ECS1 homolog 3	AAM65590.1	SEQ ID NO: 18
<i>Arabidopsis</i> ECS1 homolog 4	AAF21209.1	SEQ ID NO: 19
Rice ECS1	AK111818; BAD19260	SEQ ID NOS: 3-4
Rice ECS1 homolog 1	NM_190464; NP_915353	SEQ ID NOS: 4-5
Rice ECS1 homolog 2	NM_184451; NP_909340	SEQ ID NOS: 24-25
Rice ECS1 homolog 3	AK111801; BAD19262	SEQ ID NOS: 26-27
Rice ECS1 homolog 4	AP004069; BAD25094	SEQ ID NOS: 28-29
Rice ECS1 homolog 5	NM_197584; NP_922566	SEQ ID NOS: 30-31
Rice ECS1 homolog 6	AC051633; AAG13597	SEQ ID NOS: 32-33
Barley ECS1	Y09602; P08818, T05701	SEQ ID NOS: 7-8
Barley homolog 1	X78878; P55748	SEQ ID NOS: 9-10
Wheat ECS1	A29639	SEQ ID NO: 11
Pea ECS1	AJ251969; CAC19488	SEQ ID NOS: 12-13
<i>Medicago</i> ECS1	AY308957; AAQ63884	SEQ ID NOS: 14-15

The homology between ECS1, BRS1 and other type II serine carboxypeptidases indicated that ECS1 is a serine carboxypeptidase II-like protein. In addition, like BRS1, ECS1 is predicted to have an N terminal signal peptide and should be secreted.

As the rice genomic sequence is available, at least 5 ECS1 orthologs were first identified in rice. The alignment of the predicted amino acid sequence of ECS1 with those of the three most related rice orthologs, as well as d barley ortholog, is shown in FIG. 5.

Similar to the fact that *Arabidopsis* ECS1 has a high sequence identity compared to homologue 1 in *Arabidopsis*, but homologue 1 lacks an N-terminal signal peptide, the two rice orthologs (i.e., rice ECS1/H1 and rice ECS/H2) are identical to each other except that that rice ECS1/H2 lacks the N-terminal signal peptide as seen in rice ECS1/H1.

## EXAMPLE 4

## Expression of ECS1 and Orthologous Sequences in Selected Crop Species

The ECS1 family of genes is conserved in plants and therefore it can be predicted that overexpression of ECS1 may be used in multiple crop species to increase yield and productivity. A plan was initiated for introduction of the *Arabidopsis* ECS1 gene and identified orthologous sequences into selected crop plants including soybean, canola, maize, barley and rice. Essentially the same gene construct described above is used, consisting of a two-enhancer 35S promoter driving the ECS1 cDNA from *Arabidopsis*. Following initial expression, further studies are carried out for optimization of expression in plants grown under field conditions.

*Brassica napus* (Canola/rape) is a major oil crop closely related to *Arabidopsis*. *Agrobacterium*-mediated transforma-

tion of *Brassica* has been proven to be a routinely successful approach in recent years and therefore is the selected transformation method (Chakrabarty et al., 2002; Stewart et al., 2002). Soybeans will be transformed using the protocols described by Liu et al. (2004) and Zeng et al. (2004). Rice will be transformed with the ECS1-overexpressing construct using well known techniques (see, e.g., Lin et al., 2003; Garg et al., 2002; Wu et al., 2002; Khanna and Raina, 2002). The additional monocotyledonous species maize and barley will also be transformed using known methods for generating transgenic plants (see, e.g., Zhong et al., 1996; Horvath et al., 2003; Wan and Lemaux, 1994; Roussy et al., 2001).

Initially 10-15 transgenic plants will be obtained for each transgene (ECS1 overexpression and controls) for canola, soybean, rice and other seed crop plants. The phenotypes of the resulting T1 transgenic plants will be measured, including carpel and seed numbers, and the vegetative parts of the plants analyzed for any obvious phenotypic changes. Upon confirmation of seed yield for a given construct in the T1 ECS1 overexpressing plants, Mendelian inheritance of the phenotype will be confirmed in the T2 generation.

Following initial studies with the *Arabidopsis* ECS1 gene, optimization studies are carried out with ECS1 orthologs from other species. The rice (SEQ ID NOS:3 and 5), barley (SEQ ID NOS:7 and 9), wheat (SEQ ID NO:11), pea (SEQ ID NO:12) and *Medicago* (SEQ ID NO:14) ECS1 orthologous coding sequences are introduced. Sequences are selected for introduction into related species, such as among rice, barley and wheat.

All of the compositions and methods disclosed and claimed herein can be made and executed without undue experimentation in light of the present disclosure. While the compositions and methods of this invention have been described in terms of preferred embodiments, it will be apparent to those of skill in the art that variations may be applied to the compositions and methods and in the steps or in the sequence of steps of the method described herein without departing from the concept, spirit and scope of the invention. More specifically, it will be apparent that certain agents which are both chemically and physiologically related may be substituted for the agents described herein while the same or similar results would be achieved. All such similar substitutes and modifications apparent to those skilled in the art are deemed to be within the spirit, scope and concept of the invention as defined by the appended claims.

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U.S. Pat. No. 5,464,765  
U.S. Pat. No. 5,508,184  
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## SEQUENCE LISTING

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Ile	Tyr	Tyr	Asn	Arg	Pro	Glu	Val	Gln	Arg	Ala	Met	His	Ala	Asn	His
			325						330					335	
Thr	Ala	Ile	Pro	Tyr	Lys	Trp	Thr	Ala	Cys	Ser	Asp	Ser	Val	Phe	Asn
			340					345					350		
Asn	Trp	Asn	Trp	Arg	Asp	Ser	Asp	Asn	Ser	Met	Leu	Pro	Ile	Tyr	Lys
		355					360						365		
Glu	Leu	Ile	Ala	Ala	Gly	Leu	Arg	Ile	Trp	Val	Tyr	Ser	Gly	Asp	Thr
	370					375					380				
Asp	Ser	Val	Ile	Pro	Val	Thr	Ala	Thr	Arg	Tyr	Ser	Leu	Gly	Lys	Leu
385					390					395					400
Asn	Leu	Arg	Val	Lys	Thr	Arg	Trp	Tyr	Pro	Trp	Tyr	Ser	Gly	Asn	Gln
				405					410					415	
Val	Gly	Gly	Arg	Thr	Glu	Val	Tyr	Glu	Gly	Leu	Thr	Phe	Val	Thr	Val
			420					425					430		
Arg	Gly	Ala	Gly	His	Glu	Val	Pro	Phe	Phe	Gln	Pro	Gln	Ser	Ala	Leu
		435					440					445			
Ile	Leu	Leu	Arg	Ser	Phe	Leu	Ala	Gly	Asn	Glu	Leu	Ser	Arg	Ser	Tyr
	450					455					460				

&lt;210&gt; SEQ ID NO 3

&lt;211&gt; LENGTH: 1422

-continued

&lt;212&gt; TYPE: DNA

<213> ORGANISM: *Oryza sativa*

&lt;400&gt; SEQUENCE: 3

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atggcgacgc gagggcgat tgtagcggcg gtggcgagcg ttgtggtggc gtggctggcg      60
gtcgccgtcg gcgtaaacgg cggcggtggtc gaggcggagc gggaccgggt ggaggcgctg      120
ccggggcagc caccggtggc gttcgcgcag tacgccgggt acgtggcggt gagcgaggcg      180
agcggggcgg cgctcttcta ctggctcacc gaggcgcgcg ccgcgcgcgc cgccgccacc      240
aagccctctg tcctctgggt caacggcggt cctggatgct catcgattgc gtatggagca      300
tctgaagaga ttggccatt taggattaag acaaacggga cagggtccta tctgaacaag      360
tactcatgga acagagaggc aaacctctg ttctggaat cacctgccgg agttggcttt      420
tcatactcca acaccacctc tgatctcaag acatctggtg atgagaggac agctcaagat      480
gcgttgacgt tcttgatcag ttggatgtcc cgcttccac agtatcgga ccgggatttc      540
tacattgctg gagaagcta tctggacat tacgttcccc agttggcaag gaagatcggt      600
gagttcaaca aggcctcacc atatccttcc atcaacctca aggggatcct tgtgggcaat      660
ggggtgactg acaactacta cgacaacatc ggcacgggtg cctactggtg gacgcacgcc      720
atgatctcgg acaccaccta caaggccatc atgtcgtcgt gcaacttcac cagcgccaac      780
gtctccaggc tctgcaaccg cgccatgagc tacgccatga accacgagtt cggcgacatc      840
gaccagtaca gcattctacac gccgtctcgc gccgcgcgcg ccgcgcgcaa cgccaccggc      900
cgccgcgcgc gcaaggccgc cgtgctgagg ttcaaggaca ccttctacg gcgccggtcg      960
ttcggtacg acccctgcac ggagacatac gccgagaagt actacaaccg gccggatggt      1020
cagaaggcca tgcattgcaa catcactggg attccttaca gatggacagc ctgcagtgat      1080
gtgctcatca agacgtggcg agattcagag ttctccatgc tgccgactta caagttgctg      1140
atgaaggccg ggctgaggat atgggtgttc agtggcgaca cggattcagt cgttccgggt      1200
actgcaacga ggtttgcgct tagccatctt ggactgaaga cgaagatccg ctggtaccct      1260
tggtactcag ctggacaggt tggaggatgg tctgaggtgt atgaagggtt cacatttgcg      1320
tcagtgaagc gtgctgggca tgaggtgcca ctgtttcagc caaggagagc attcaggatg      1380
tttcagtcgt tcttggcagg ggagccattg ccaaaatcct ga                          1422

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&lt;210&gt; SEQ ID NO 4

&lt;211&gt; LENGTH: 473

&lt;212&gt; TYPE: PRT

<213> ORGANISM: *Oryza sativa*

&lt;400&gt; SEQUENCE: 4

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Met Ala Thr Arg Gly Arg Ile Val Ala Ala Val Ala Ser Val Val Val
 1             5             10            15
Ala Trp Leu Ala Val Ala Val Gly Val Asn Gly Gly Gly Cys Glu Ala
 20            25            30
Glu Arg Asp Arg Val Glu Ala Leu Pro Gly Gln Pro Pro Val Ala Phe
 35            40            45
Ala Gln Tyr Ala Gly Tyr Val Ala Val Ser Glu Ala Ser Gly Arg Ala
 50            55            60
Leu Phe Tyr Trp Leu Thr Glu Ala Ala Ala Ala Ala Ala Ala Thr
 65            70            75            80
Lys Pro Leu Val Leu Trp Leu Asn Gly Gly Pro Gly Cys Ser Ser Ile
 85            90            95

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Ala	Tyr	Gly	Ala	Ser	Glu	Glu	Ile	Gly	Pro	Phe	Arg	Ile	Lys	Thr	Asn
			100					105					110		
Gly	Thr	Gly	Leu	Tyr	Leu	Asn	Lys	Tyr	Ser	Trp	Asn	Arg	Glu	Ala	Asn
		115					120					125			
Leu	Leu	Phe	Leu	Glu	Ser	Pro	Ala	Gly	Val	Gly	Phe	Ser	Tyr	Ser	Asn
		130				135					140				
Thr	Thr	Ser	Asp	Leu	Lys	Thr	Ser	Gly	Asp	Glu	Arg	Thr	Ala	Gln	Asp
145					150					155					160
Ala	Leu	Gln	Phe	Leu	Ile	Ser	Trp	Met	Ser	Arg	Phe	Pro	Gln	Tyr	Arg
				165					170					175	
His	Arg	Asp	Phe	Tyr	Ile	Ala	Gly	Glu	Ser	Tyr	Ala	Gly	His	Tyr	Val
			180					185					190		
Pro	Gln	Leu	Ala	Arg	Lys	Ile	Val	Glu	Phe	Asn	Lys	Ala	Ser	Pro	Tyr
		195					200					205			
Pro	Phe	Ile	Asn	Leu	Lys	Gly	Ile	Leu	Val	Gly	Asn	Gly	Val	Thr	Asp
	210					215					220				
Asn	Tyr	Tyr	Asp	Asn	Ile	Gly	Thr	Val	Thr	Tyr	Trp	Trp	Thr	His	Ala
225					230					235					240
Met	Ile	Ser	Asp	Thr	Thr	Tyr	Lys	Ala	Ile	Met	Ser	Ser	Cys	Asn	Phe
				245					250					255	
Thr	Ser	Ala	Asn	Val	Ser	Arg	Leu	Cys	Asn	Arg	Ala	Met	Ser	Tyr	Ala
			260					265					270		
Met	Asn	His	Glu	Phe	Gly	Asp	Ile	Asp	Gln	Tyr	Ser	Ile	Tyr	Thr	Pro
		275					280					285			
Ser	Cys	Ala	Ala	Ala	Ala	Ala	Ala	Asn	Ala	Thr	Gly	Arg	Arg	Arg	Gly
	290					295					300				
Lys	Ala	Ala	Val	Leu	Arg	Phe	Lys	Asp	Thr	Phe	Leu	Arg	Arg	Arg	Ser
305					310					315					320
Phe	Gly	Tyr	Asp	Pro	Cys	Thr	Glu	Thr	Tyr	Ala	Glu	Lys	Tyr	Tyr	Asn
				325					330					335	
Arg	Pro	Asp	Val	Gln	Lys	Ala	Met	His	Ala	Asn	Ile	Thr	Gly	Ile	Pro
			340					345					350		
Tyr	Arg	Trp	Thr	Ala	Cys	Ser	Asp	Val	Leu	Ile	Lys	Thr	Trp	Arg	Asp
			355				360					365			
Ser	Glu	Phe	Ser	Met	Leu	Pro	Thr	Tyr	Lys	Leu	Leu	Met	Lys	Ala	Gly
	370					375					380				
Leu	Arg	Ile	Trp	Val	Phe	Ser	Gly	Asp	Thr	Asp	Ser	Val	Val	Pro	Val
385					390					395				400	
Thr	Ala	Thr	Arg	Phe	Ala	Leu	Ser	His	Leu	Gly	Leu	Lys	Thr	Lys	Ile
				405					410					415	
Arg	Trp	Tyr	Pro	Trp	Tyr	Ser	Ala	Gly	Gln	Val	Gly	Gly	Trp	Ser	Glu
			420					425					430		
Val	Tyr	Glu	Gly	Leu	Thr	Phe	Ala	Ser	Val	Arg	Gly	Ala	Gly	His	Glu
		435					440					445			
Val	Pro	Leu													

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<210> SEQ ID NO 5
<211> LENGTH: 2010
<212> TYPE: DNA
<213> ORGANISM: Oryza sativa

<400> SEQUENCE: 5
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atggccatca gtagcagagc agctgcgtgc ggcgcgctca tcttcccgac caccgcatcc	60
gccgtctccg tctcccgag cgtctccgtg gaccaaagag tcagccaccg gcggagggaag	120
gcggtggcgg tggcggccgt gccgcacgcc agcagcggcg gcgcgctgct ggagcggccg	180
gccttcgacc agtcccagct cgacacgctt cccgtgacac aagaaggagg ggacaccgga	240
aggatgaggg acaggagggg ctctggaagc ggtgacagct acaaagtttt gctcatagac	300
gacgcccgcc acaccagaa gcttgtggag aaggccttgc cgcagggtgt gccgtccgtg	360
accgcggagg cggcgcggca gctcttccac gcgtcccgcc agaaaggcgc cgcgctcgtc	420
attgtcgccg tgaagcttct tctacctccg ttccacacgc gcgcctcgc tcgccgccag	480
cgccgccgcc accaccacca ccgccactgc cactatacta atgccgagtt gccgacacc	540
ccacttgccc cgccgcgtcg ctgcgtaca gcgctagagc gagctagcac actagcagtg	600
agccagtgtc ccgtggtccg gccattggag attttggagc tcgtaaatggc tcacaaggcc	660
gcggtctctg tgetgctgct agtgtcagtg tcagtggcgg ccgcggcgctc gggcgaccag	720
gagagcgacc ggatccggga gctccccggg cagccggcga aggtgaggtt ctgcagtac	780
tccggtctac tgacggctca ccaggcgac gcccgcgccg tcttctactg gctggtggag	840
gcggtgcggc cggccggggc catcgcgccg ctgcctcgtt ggctcaacgg cgggcggggg	900
tgctcgtcgg tcgggtacgg cgcgtcggag gaggtcggcc cgttcgggat caggcccgac	960
gggaagacgc tgtacctgaa cccaattct tggaacaagg cggcgaattt gctgttcttg	1020
gagtcgccgg ccggcgtggg gttctcgtac tcgaacaaga cgttggatct gtacgtcgca	1080
ggagatgcta agacagcatc ggatgcttat gcatttctgg tgaactgggt ggagagattc	1140
ccacaataca agtacaggga gttctacatt gctggggaga gctatgcagg gcattacgtt	1200
ccccagttag ccagctcat ctatgaacag aacaagggca ttcagaatcc aataattaat	1260
ctcaaaggat tcattggtggg taatgcggtt actgatgact accacgacta tcttggtacc	1320
tttgagtatt ggtggactca tggcctcctc tctgacaaca cttatcaca cctgaagaag	1380
acatgcttgc ttgagtcctc tgagcaccct tctcctgaat gtctaaagaa cctgaacctc	1440
gccagttcag aagaaggcaa tatcgatcct tacagcctgt atacaaagcc ctgcaataat	1500
acagcctctc tcaaacttgg cttgggagga cgctaccctt gggtatccag agcatatgat	1560
ccctgcacag aaagatactc aagtatttac tacaaccggc cagaagtgca gatagcgatg	1620
catgctaaca ccactgggat tcaatattca tggaaaactt gcagcgatat tgcggatca	1680
tactgggcag attccccgaa atctatgctt cctatctacc aagaattgat tgcagctggt	1740
atcaggatat gggttttcag tggggataca gatgctgtag ttctgttac tgcaacaagg	1800
tactcaatag atgctcttaa gcttccaact atgggtcaatt ggtacccttg gtatgaccac	1860
ggaaagggtg gaggttgag tcaagtgtat aaaggattaa ctctcgtcac tatagcaggc	1920
gcaggccatg aggtaccact acaccggcct cgagaagcac ttatattatt cagacacttc	1980
ttgcagaata caccatgcc aactcaatag	2010

&lt;210&gt; SEQ ID NO 6

&lt;211&gt; LENGTH: 669

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Oryza sativa

&lt;400&gt; SEQUENCE: 6

Met Ala Ile Ser Ser Arg Ala Ala Ala Cys Gly Ala Leu Ile Phe Pro  
 1 5 10 15



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Thr	Thr	Ala	Ser	Ala	Ala	Pro	Val	Ser	Arg	Ser	Val	Ser	Val	Asp	Gln
			20					25					30		
Arg	Val	Ser	His	Arg	Arg	Arg	Lys	Ala	Val	Ala	Val	Ala	Ala	Val	Pro
		35					40					45			
His	Ala	Ser	Ser	Gly	Gly	Ala	Leu	Leu	Glu	Arg	Pro	Ala	Phe	Asp	Gln
	50					55					60				
Ser	Gln	Leu	Asp	Thr	Leu	Pro	Val	Thr	Gln	Glu	Gly	Gly	Asp	Thr	Gly
	65				70					75					80
Arg	Met	Arg	Asp	Arg	Arg	Gly	Ser	Gly	Ser	Gly	Asp	Ser	Tyr	Lys	Val
			85						90					95	
Leu	Leu	Ile	Asp	Asp	Ala	Arg	His	Thr	Glu	Lys	Leu	Val	Glu	Lys	Ala
			100					105					110		
Leu	Pro	Gln	Val	Val	Pro	Ser	Val	Thr	Ala	Glu	Ala	Ala	Arg	Gln	Leu
		115					120					125			
Phe	His	Ala	Ser	Arg	Gln	Lys	Gly	Ala	Ala	Leu	Val	Ile	Val	Ala	Val
	130					135					140				
Lys	Leu	Leu	Leu	Pro	Pro	Phe	His	Thr	Arg	Ala	Leu	Ala	Arg	Arg	Gln
	145				150					155					160
Arg	Arg	Arg	His	His	His	His	Arg	His	Cys	His	Tyr	Thr	Asn	Ala	Glu
			165						170					175	
Leu	Pro	Thr	Pro	Pro	Leu	Ala	Pro	Pro	Arg	Arg	Cys	Ala	Thr	Ala	Leu
			180					185					190		
Glu	Arg	Ala	Ser	Thr	Leu	Ala	Val	Ser	Gln	Cys	Pro	Val	Val	Arg	Pro
		195					200					205			
Leu	Glu	Ile	Leu	Glu	Leu	Val	Met	Ala	His	Lys	Ala	Ala	Ala	Leu	Val
	210					215					220				
Leu	Leu	Leu	Val	Ser	Val	Ser	Val	Ala	Ala	Ala	Ala	Ser	Gly	Asp	Gln
	225				230					235					240
Glu	Ser	Asp	Arg	Ile	Arg	Glu	Leu	Pro	Gly	Gln	Pro	Ala	Lys	Val	Arg
			245						250					255	
Phe	Ser	Gln	Tyr	Ser	Gly	Tyr	Val	Thr	Val	Asn	Gln	Ala	His	Gly	Arg
			260					265					270		
Ala	Leu	Phe	Tyr	Trp	Leu	Val	Glu	Ala	Val	Pro	Ala	Ala	Gly	Pro	Ile
		275					280					285			
Ala	Pro	Leu	Val	Leu	Trp	Leu	Asn	Gly	Gly	Pro	Gly	Cys	Ser	Ser	Val
		290				295					300				
Gly	Tyr	Gly	Ala	Ser	Glu	Glu	Val	Gly	Pro	Phe	Arg	Ile	Arg	Pro	Asp
	305				310					315					320
Gly	Lys	Thr	Leu	Tyr	Leu	Asn	Pro	Asn	Ser	Trp	Asn	Lys	Ala	Ala	Asn
			325						330					335	
Leu	Leu	Phe	Leu	Glu	Ser	Pro	Ala	Gly	Val	Gly	Phe	Ser	Tyr	Ser	Asn
		340						345					350		
Lys	Thr	Leu	Asp	Leu	Tyr	Val	Ala	Gly	Asp	Ala	Lys	Thr	Ala	Ser	Asp
		355					360					365			
Ala	Tyr	Ala	Phe	Leu	Val	Asn	Trp	Leu	Glu	Arg	Phe	Pro	Gln	Tyr	Lys
		370				375					380				
Tyr	Arg	Glu	Phe	Tyr	Ile	Ala	Gly	Glu	Ser	Tyr	Ala	Gly	His	Tyr	Val
	385				390					395					400
Pro	Gln	Leu	Ala	Gln	Leu	Ile	Tyr	Glu	Gln	Asn	Lys	Gly	Ile	Gln	Asn
			405						410					415	
Pro	Ile	Ile	Asn	Leu	Lys	Gly	Phe	Met	Val	Gly	Asn	Ala	Val	Thr	Asp
			420					425						430	

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Asp	Tyr	His	Asp	Tyr	Leu	Gly	Thr	Phe	Glu	Tyr	Trp	Trp	Thr	His	Gly
		435					440					445			
Leu	Ile	Ser	Asp	Asn	Thr	Tyr	His	Asn	Leu	Lys	Lys	Thr	Cys	Leu	Leu
	450					455					460				
Glu	Ser	Ser	Glu	His	Pro	Ser	Pro	Glu	Cys	Leu	Lys	Asn	Leu	Asn	Leu
465					470					475				480	
Ala	Ser	Ser	Glu	Glu	Gly	Asn	Ile	Asp	Pro	Tyr	Ser	Leu	Tyr	Thr	Lys
			485					490						495	
Pro	Cys	Asn	Asn	Thr	Ala	Ser	Leu	Lys	Leu	Gly	Leu	Gly	Gly	Arg	Tyr
		500						505					510		
Pro	Trp	Leu	Ser	Arg	Ala	Tyr	Asp	Pro	Cys	Thr	Glu	Arg	Tyr	Ser	Ser
	515						520					525			
Ile	Tyr	Tyr	Asn	Arg	Pro	Glu	Val	Gln	Ile	Ala	Met	His	Ala	Asn	Thr
	530					535					540				
Thr	Gly	Ile	Gln	Tyr	Ser	Trp	Lys	Thr	Cys	Ser	Asp	Ile	Val	Gly	Ser
545					550					555				560	
Tyr	Trp	Ala	Asp	Ser	Pro	Lys	Ser	Met	Leu	Pro	Ile	Tyr	Gln	Glu	Leu
			565					570						575	
Ile	Ala	Ala	Gly	Ile	Arg	Ile	Trp	Val	Phe	Ser	Gly	Asp	Thr	Asp	Ala
		580						585					590		
Val	Val	Pro	Val	Thr	Ala	Thr	Arg	Tyr	Ser	Ile	Asp	Ala	Leu	Lys	Leu
		595					600				605				
Pro	Thr	Met	Val	Asn	Trp	Tyr	Pro	Trp	Tyr	Asp	His	Gly	Lys	Val	Gly
	610					615				620					
Gly	Trp	Ser	Gln	Val	Tyr	Lys	Gly	Leu	Thr	Leu	Val	Thr	Ile	Ala	Gly
625					630					635				640	
Ala	Gly	His	Glu	Val	Pro	Leu	His	Arg	Pro	Arg	Glu	Ala	Leu	Ile	Leu
			645					650						655	
Phe	Arg	His	Phe	Leu	Gln	Asn	Thr	Pro	Met	Pro	Thr	Gln			
		660						665							

&lt;210&gt; SEQ ID NO 7

&lt;211&gt; LENGTH: 1431

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Hordeum vulgare

&lt;400&gt; SEQUENCE: 7

atgaggacta cgaccgcgcg tctcccccca gctccggcgg cggcggcggt gctcctggcg	60
gcgttgacgt gcctcctcct ccggccagcc gccgtgcgcg cggcgggcgg ccattgccgcg	120
gaccgcatag tccggctgcc ggggcagccg gaggtggact tcgacatgta ctccgggtac	180
atcacggtgg acgaggccgc cggacggtcg ctcttctacc tgctgcagga ggcgcccag	240
gaggccacgc cggcgccgct cgtgctgtgg ctcaacggcg gcccggctg ctctccgctc	300
gcctacggcg cgtcggagga gctcggcgcg ttccgcgtca tgccccgcgg cgccggcctc	360
gtcctcaacg agtaccgctg gaacaaagtg gccaacgtgc tgttctgga ttgcgcggcc	420
ggcgtggggg tctcctacac caaccacgc tccgacatct acacctccgg cgacaacagg	480
acggcgcaag actcgtacgc ctctcctggcg gcatgggtcg agaggttccc gactacaag	540
taccgcgaat tctacgtcgc cggcgagagc tacgcggggc actacgtccc ggagctgtcg	600
cagctgggtcc accggagcgg caaccctgc atcaacctca agggcttcat ggtcggcaac	660
ggcctcatcg acgactacca cgactacgtc ggcaccttcg agttctgtgtg gaaccacggg	720
atcgtctccg acgacaccta ccgccgcctc aaggacgcct gcctccacga ctcttctcat	780

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cacccctcgc cggcgtgcga cgcgcgcgcg gacgtcgcca cggcggagca gggcaacatc	840
gacatgtaca gcctctacac ccccgctctgc aacatctcgt cgtcgtcgtc gtcgtcgtcc	900
ttgagccggc ggcggaccag agggcgctac ccatggctga cggggtcgta cgaccctgtc	960
acggagaggt actcgacggc gtactacaac cggcgggacg tgcagacggc cctccacgcc	1020
aacgtcaccg gcgccatgaa ctacacgtgg gcgacctgca gcgacccat taatacccac	1080
tggcatgatg ctccgaggtc catgcttccc atctacaggg agctgattgc agctggccta	1140
aggatttggg tcttcagcgg cgacacggat gcggtagtcc ccttgacagc aacaagatac	1200
tccatcggcg ctctgggtct tgcaactact accagttggt acccttggtg tgacgacctg	1260
caggaggtcg gcgctggag ccaggtgtac aagggcctta cgctggtgtc cgtcagaggt	1320
gcgggccatg aggttctctc gcaccgtccg cggcaagcgc tcatactgtt tcagcaattc	1380
ctgcagggca agcccatgcc aggccgtacc acaaatgtga cggtggctta a	1431

&lt;210&gt; SEQ ID NO 8

&lt;211&gt; LENGTH: 476

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Hordeum vulgare

&lt;400&gt; SEQUENCE: 8

Met Arg Thr Thr Thr Arg Arg Leu Pro Pro Ala Pro Ala Ala Ala Ala	1 5 10 15
Val Leu Leu Ala Ala Leu Thr Cys Leu Leu Leu Arg Pro Ala Ala Val	20 25 30
Ala Ala Ala Gly Gly His Ala Ala Asp Arg Ile Val Arg Leu Pro Gly	35 40 45
Gln Pro Glu Val Asp Phe Asp Met Tyr Ser Gly Tyr Ile Thr Val Asp	50 55 60
Glu Ala Ala Gly Arg Ser Leu Phe Tyr Leu Leu Gln Glu Ala Pro Glu	65 70 75 80
Glu Ala Gln Pro Ala Pro Leu Val Leu Trp Leu Asn Gly Gly Pro Gly	85 90 95
Cys Ser Ser Val Ala Tyr Gly Ala Ser Glu Glu Leu Gly Ala Phe Arg	100 105 110
Val Met Pro Arg Gly Ala Gly Leu Val Leu Asn Glu Tyr Arg Trp Asn	115 120 125
Lys Val Ala Asn Val Leu Phe Leu Asp Ser Pro Ala Gly Val Gly Phe	130 135 140
Ser Tyr Thr Asn Thr Ser Ser Asp Ile Tyr Thr Ser Gly Asp Asn Arg	145 150 155 160
Thr Ala His Asp Ser Tyr Ala Phe Leu Ala Ala Trp Phe Glu Arg Phe	165 170 175
Pro His Tyr Lys Tyr Arg Glu Phe Tyr Val Ala Gly Glu Ser Tyr Ala	180 185 190
Gly His Tyr Val Pro Glu Leu Ser Gln Leu Val His Arg Ser Gly Asn	195 200 205
Pro Val Ile Asn Leu Lys Gly Phe Met Val Gly Asn Gly Leu Ile Asp	210 215 220
Asp Tyr His Asp Tyr Val Gly Thr Phe Glu Phe Trp Trp Asn His Gly	225 230 235 240
Ile Val Ser Asp Asp Thr Tyr Arg Arg Leu Lys Asp Ala Cys Leu His	245 250 255
Asp Ser Phe Ile His Pro Ser Pro Ala Cys Asp Ala Ala Thr Asp Val	

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260	265	270
Ala Thr Ala Glu Gln Gly Asn Ile Asp Met Tyr Ser Leu Tyr Thr Pro 275 280 285		
Val Cys Asn Ile Ser Ser Ser Ser Ser Ser Ser Ser Leu Ser Arg Arg 290 295 300		
Arg Thr Arg Gly Arg Tyr Pro Trp Leu Thr Gly Ser Tyr Asp Pro Cys 305 310 315 320		
Thr Glu Arg Tyr Ser Thr Ala Tyr Tyr Asn Arg Arg Asp Val Gln Thr 325 330 335		
Ala Leu His Ala Asn Val Thr Gly Ala Met Asn Tyr Thr Trp Thr Asn 340 345 350		
Cys Ser Asp Thr Ile Asn Thr His Trp His Asp Ala Pro Arg Ser Met 355 360 365		
Leu Pro Ile Tyr Arg Glu Leu Ile Ala Ala Gly Leu Arg Ile Trp Val 370 375 380		
Phe Ser Gly Asp Thr Asp Ala Val Val Pro Leu Thr Ala Thr Arg Tyr 385 390 395 400		
Ser Ile Gly Ala Leu Gly Leu Ala Thr Thr Thr Ser Trp Tyr Pro Trp 405 410 415		
Tyr Asp Asp Leu Gln Glu Val Gly Gly Trp Ser Gln Val Tyr Lys Gly 420 425 430		
Leu Thr Leu Val Ser Val Arg Gly Ala Gly His Glu Val Pro Leu His 435 440 445		
Arg Pro Arg Gln Ala Leu Ile Leu Phe Gln Gln Phe Leu Gln Gly Lys 450 455 460		
Pro Met Pro Gly Arg Thr Thr Asn Val Thr Val Ala 465 470 475		

&lt;210&gt; SEQ ID NO 9

&lt;211&gt; LENGTH: 1580

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Hordeum vulgare

&lt;400&gt; SEQUENCE: 9

```

cgggtgccgcg ggtgccggg caggccttcg acgccagctt cgcgcactac gccggctacg      60
tcaccgtcag cgaggaccgc ggcgcgcgcg tcttctactg gttcttcgag gccgcgcacg      120
acccggcctc caagccgctc ctgctctggc tcaacggagg gcctggttgc tcatcgattg      180
cttttgagtg cggggaagaa gtggggcctt tccatgtcaa tgcagacgga aagggcggtc      240
atatgaatcc ttactcttgg aaccaagttg caaatatctt gttccttgat tcaccggttg      300
gtgttggtta ttcatttca aacacctctg ctgatatttt aagcaatggg gatgagagga      360
ctgccaagga ttcgttggtg ttctaacaa agtggcttga acgattccct caatacaagg      420
agcgtgaatt ttatttaact ggagagagct atgctggaca ctacgttctc cagttggctc      480
aagccataaa gaggcacatc gaggccactg gagacaaatc aatcaatcta aaggggttata      540
tggtaggaaa tgccctgact gacgatttcc atgaccacta tggaatatat caatatatgt      600
ggaccactgg cttgatttct gatcaaacat acaagctact gaacattttc tgtgacttcg      660
agtcctttgt gcatacatct ccacagtgtg ataagattct tgacattgct agcactgaag      720
ctgggaacat tgattcgtat agcatcttca cacctacttg tcattcatct tttgcctcct      780
caaggaacaa agtggtgaaa aggccttcgg ctgttggaat aatgggggag caatacgatc      840
catgtaccga aaaacattca attgtatatt tcaatctgca tgagggtgcag aaggcacttc      900

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acgtcaatcc ggtcattggc aaatccaaat gggagacctg cagtgaagtt attaacacca 960
actggaagga ctgtgaaaga tctgtattgc atatctatca tgaacttatt cagtatgggc 1020
ttcgtatatg gatgttcagt ggagacacag atgcagtgat tccagtaaca tcaactagat 1080
acagcattga tgctctcaag ctccaacag tgaccccgctg gcatgcttgg tatgatgatg 1140
atggcgaggt tgggtggttg acccaagggt acaagggtct caactttgtg acagtaaggg 1200
gtgcggttca tgaggttcct ctccatcgct ccaagcaggc tcttacgctc atcaaatcat 1260
tcttgcccg gaggccaatg cctgtgctgt ctgatctacg cagcgatatg taatatgccg 1320
gacacatttg gtttcggaca cgaccagcac cacaagattc cagctacca aggcagttcg 1380
gttggttaaaa ctccacacgt acttcacaaa tataaggatg gccatagctg ttgccatttg 1440
taagtgtat tggcaccaat taatcccgty agacaggga acagttttcc tggcgcta 1500
tgacactgca gcactgcctg ttaaattaat ctggaactaa ggataaagat gaattgaatt 1560
tccccaaaaa aaaaaaaaaa 1580

```

&lt;210&gt; SEQ ID NO 10

&lt;211&gt; LENGTH: 436

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Hordeum vulgare

&lt;400&gt; SEQUENCE: 10

```

Val Pro Arg Val Pro Gly Gln Ala Phe Asp Ala Ser Phe Ala His Tyr
  1             5             10             15
Ala Gly Tyr Val Thr Val Ser Glu Asp Arg Gly Ala Ala Leu Phe Tyr
             20             25             30
Trp Phe Phe Glu Ala Ala His Asp Pro Ala Ser Lys Pro Leu Leu Leu
             35             40             45
Trp Leu Asn Gly Gly Pro Gly Cys Ser Ser Ile Ala Phe Gly Val Gly
             50             55             60
Glu Glu Val Gly Pro Phe His Val Asn Ala Asp Gly Lys Gly Val His
             65             70             75             80
Met Asn Pro Tyr Ser Trp Asn Gln Val Ala Asn Ile Leu Phe Leu Asp
             85             90             95
Ser Pro Val Gly Val Gly Tyr Ser Tyr Ser Asn Thr Ser Ala Asp Ile
             100            105            110
Leu Ser Asn Gly Asp Glu Arg Thr Ala Lys Asp Ser Leu Val Phe Leu
             115            120            125
Thr Lys Trp Leu Glu Arg Phe Pro Gln Tyr Lys Glu Arg Glu Phe Tyr
             130            135            140
Leu Thr Gly Glu Ser Tyr Ala Gly His Tyr Val Pro Gln Leu Ala Gln
             145            150            155            160
Ala Ile Lys Arg His His Glu Ala Thr Gly Asp Lys Ser Ile Asn Leu
             165            170            175
Lys Gly Tyr Met Val Gly Asn Ala Leu Thr Asp Asp Phe His Asp His
             180            185            190
Tyr Gly Ile Phe Gln Tyr Met Trp Thr Thr Gly Leu Ile Ser Asp Gln
             195            200            205
Thr Tyr Lys Leu Leu Asn Ile Phe Cys Asp Phe Glu Ser Phe Val His
             210            215            220
Thr Ser Pro Gln Cys Asp Lys Ile Leu Asp Ile Ala Ser Thr Glu Ala
             225            230            235            240
Gly Asn Ile Asp Ser Tyr Ser Ile Phe Thr Pro Thr Cys His Ser Ser
             245            250            255

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Phe Ala Ser Ser Arg Asn Lys Val Val Lys Arg Leu Arg Ser Val Gly  
                   260                  265                  270  
 Lys Met Gly Glu Gln Tyr Asp Pro Cys Thr Glu Lys His Ser Ile Val  
                   275                  280                  285  
 Tyr Phe Asn Leu His Glu Val Gln Lys Ala Leu His Val Asn Pro Val  
                   290                  295                  300  
 Ile Gly Lys Ser Lys Trp Glu Thr Cys Ser Glu Val Ile Asn Thr Asn  
                   305                  310                  315                  320  
 Trp Lys Asp Cys Glu Arg Ser Val Leu His Ile Tyr His Glu Leu Ile  
                   325                  330                  335  
 Gln Tyr Gly Leu Arg Ile Trp Met Phe Ser Gly Asp Thr Asp Ala Val  
                   340                  345                  350  
 Ile Pro Val Thr Ser Thr Arg Tyr Ser Ile Asp Ala Leu Lys Leu Pro  
                   355                  360                  365  
 Thr Val Thr Pro Trp His Ala Trp Tyr Asp Asp Asp Gly Glu Val Gly  
                   370                  375                  380  
 Gly Trp Thr Gln Gly Tyr Lys Gly Leu Asn Phe Val Thr Val Arg Gly  
                   385                  390                  395                  400  
 Ala Gly His Glu Val Pro Leu His Arg Pro Lys Gln Ala Leu Thr Leu  
                   405                  410                  415  
 Ile Lys Ser Phe Leu Ala Gly Arg Pro Met Pro Val Leu Ser Asp Leu  
                   420                  425                  430  
 Arg Ser Asp Met  
                   435  
  
 <210> SEQ ID NO 11  
 <211> LENGTH: 423  
 <212> TYPE: PRT  
 <213> ORGANISM: Triticum aestivum  
  
 <400> SEQUENCE: 11  
  
 Val Glu Pro Ser Gly His Ala Ala Asp Arg Ile Ala Arg Leu Pro Gly  
   1                  5                  10                  15  
 Gln Pro Ala Val Asp Phe Asp Met Tyr Ser Gly Tyr Ile Thr Val Asp  
                   20                  25                  30  
 Glu Gly Ala Gly Arg Ser Leu Phe Tyr Leu Leu Gln Glu Ala Pro Glu  
                   35                  40                  45  
 Asp Ala Gln Pro Ala Pro Leu Val Leu Trp Leu Asn Gly Gly Pro Gly  
                   50                  55                  60  
 Cys Ser Ser Val Ala Tyr Gly Ala Ser Glu Glu Leu Gly Ala Phe Arg  
                   65                  70                  75                  80  
 Val Lys Pro Arg Gly Ala Gly Leu Val Leu Asn Glu Tyr Arg Trp Asn  
                   85                  90                  95  
 Lys Val Ala Asn Val Leu Phe Leu Asp Ser Pro Ala Gly Val Gly Phe  
                   100                  105                  110  
 Ser Tyr Thr Asn Thr Ser Ser Asp Ile Tyr Thr Ser Gly Asp Asn Arg  
                   115                  120                  125  
 Thr Ala His Asp Ser Tyr Ala Phe Leu Ala Lys Trp Phe Glu Arg Phe  
                   130                  135                  140  
 Pro His Tyr Lys Tyr Arg Asp Phe Tyr Ile Ala Gly Glu Ser Tyr Ala  
                   145                  150                  155                  160  
 Gly His Tyr Val Pro Glu Leu Ser Gln Leu Val His Arg Ser Lys Asn  
                   165                  170                  175  
 Pro Val Ile Asn Leu Lys Gly Phe Met Val Gly Asn Gly Leu Ile Asp

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180							185					190				
Asp	Tyr	His	Asp	Tyr	Val	Gly	Thr	Phe	Glu	Phe	Trp	Trp	Asn	His	Gly	
		195					200					205				
Ile	Val	Ser	Asp	Asp	Thr	Tyr	Arg	Arg	Leu	Lys	Glu	Ala	Cys	Leu	His	
	210					215					220					
Asp	Ser	Phe	Ile	His	Pro	Ser	Pro	Ala	Cys	Asp	Ala	Ala	Thr	Asp	Val	
225					230					235					240	
Ala	Thr	Ala	Glu	Gln	Gly	Asn	Ile	Asp	Met	Tyr	Ser	Leu	Tyr	Thr	Pro	
				245					250					255		
Val	Cys	Asn	Ile	Thr	Ser	Ser	Thr	Gly	Ser	Tyr	Asp	Pro	Cys	Thr	Glu	
			260					265					270			
Arg	Tyr	Ser	Thr	Ala	Tyr	Tyr	Asn	Arg	Arg	Asp	Val	Gln	Met	Ala	Leu	
		275					280					285				
His	Ala	Asn	Val	Thr	Gly	Ala	Met	Asn	Tyr	Thr	Trp	Ala	Thr	Cys	Ser	
	290					295					300					
Asp	Thr	Ile	Asn	Thr	His	Trp	His	Asp	Ala	Pro	Arg	Ser	Met	Leu	Pro	
305					310					315					320	
Ile	Tyr	Arg	Glu	Leu	Ile	Ala	Ala	Gly	Leu	Arg	Ile	Trp	Val	Phe	Ser	
				325					330					335		
Gly	Asp	Thr	Asp	Ala	Val	Val	Pro	Leu	Thr	Ala	Thr	Arg	Tyr	Ser	Ile	
			340					345					350			
Gly	Ala	Leu	Gly	Leu	Pro	Thr	Thr	Ser	Trp	Tyr	Pro	Trp	Tyr	Asp		
	355					360					365					
Asp	Gln	Glu	Val	Gly	Gly	Trp	Ser	Gln	Val	Tyr	Lys	Gly	Leu	Thr	Leu	
	370					375					380					
Val	Ser	Val	Arg	Gly	Ala	Gly	His	Glu	Val	Pro	Leu	His	Arg	Pro	Arg	
	385				390					395					400	
Gln	Ala	Leu	Val	Leu	Phe	Gln	Tyr	Phe	Leu	Gln	Gly	Lys	Pro	Met	Pro	
				405					410					415		
Gly	Gln	Thr	Lys	Asn	Ala	Thr										
			420													

&lt;210&gt; SEQ ID NO 12

&lt;211&gt; LENGTH: 1485

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Pisum sativum

&lt;400&gt; SEQUENCE: 12

```

gaaacttctc ttcttctatc ctttctcatt attctctcac actttgtggt tgaaatccat      60
ggaaaaaaca aacaagtga agctcttgac aatcttcaca aagcagaata catagaaaat      120
tcagaaattg ataagagtga atttgaagta caagagattg tgtatgacat tgatgccatt      180
gctgattctc aaaaggtgtg caaagagaat gatagaatca aaaagcttcc tggtaacccc      240
tttgtgaaat tctctcaatt tggagggtat gttacattgg ataaattgag tggtagtgcg      300
ttttactatt actttgttga agctcatcaa tctaagaaa cacctccact tcttctttgg      360
ctcaatggag gtctgtgatg ttcattctta gcttatggag caatgcaaga attgggacct      420
tttagagtaa acagtgatgg caaaacactt caccaaaata gatactcatg gaattatgct      480
gcaaatgttt tgttcttgga gtctccagtt ggagtaggat tttcttactc aaacaaatca      540
acagaatata gtagcaatgg agacaagaaa acagctatag ataactattt atttttggtg      600
aattggttgg aaagatttcc agaataataa aatagagatt tttatatttc tggagaaagc      660
tatgctggac attatgttcc tcaacttgca cataccatcc tctatcataa taaaaaggca      720

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aataaaacaa tcattaacct caaaggaatc ttgatagga atgcagtgat ccatgatact 780
acagactcaa caggaatgta tgattttctt gctactcatg ctatcatctc agacaaagca 840
gcttatgatg tcaacaaagt ttgcgatttc tcgtcatcag ataatctcac tgctgaatgc 900
aattcagctg ctgatgaagt taatgaagat attgcattca tcgatttgta taacatttat 960
gctccactat gcaagaatga gaatctcact tccaagccca aaaagaacac tattgtgact 1020
gatccatgca gtaagaatta tgtgtatgct tatcttaata gacaagatgt tcaagaggct 1080
attcatgcta atgtcacaaa actcaaatat gaatggagtc catgcagtggtgtcattaga 1140
aaatgggttg atagctctcc aacagttctt cctcttttac atgaattcct caataatggc 1200
cttagagttt ggattttcag cggtgacacg gatggaagggttctctgttac ttcgactaag 1260
tattcgatta agaagatgaa ccttctctgtt aaaactgttt ggcacccttg gttcgccctat 1320
ggagaagttg gtggctatac tgaagtatac aaggagagacc taacatttgt tacagtgaga 1380
gaagcaggac atcaagtgcc aagttatcaa ccagcaagag ctcttacttt gattaaacat 1440
ttcttgatg gcactctctt tccttctcca aaaataaaag catag 1485

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&lt;210&gt; SEQ ID NO 13

&lt;211&gt; LENGTH: 494

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Pisum sativum

&lt;400&gt; SEQUENCE: 13

```

Glu Thr Ser Leu Leu Ser Phe Leu Ile Ile Leu Ser His Phe Val
 1             5             10             15
Val Glu Ile His Gly Lys Asn Lys Gln Val Glu Ala Leu Asp Asn Leu
      20             25             30
His Lys Ala Glu Tyr Ile Glu Asn Ser Glu Ile Asp Lys Ser Glu Phe
      35             40             45
Glu Val Gln Glu Ile Val Tyr Asp Ile Asp Ala Ile Ala Asp Ser Gln
      50             55             60
Lys Gly Val Lys Glu Asn Asp Arg Ile Lys Lys Leu Pro Gly Gln Pro
      65             70             75             80
Phe Val Lys Phe Ser Gln Phe Gly Gly Tyr Val Thr Leu Asp Lys Leu
      85             90             95
Ser Gly Ser Ala Phe Tyr Tyr Tyr Phe Val Glu Ala His Gln Ser Lys
      100            105            110
Glu Thr Pro Pro Leu Leu Leu Trp Leu Asn Gly Gly Pro Gly Cys Ser
      115            120            125
Ser Leu Ala Tyr Gly Ala Met Gln Glu Leu Gly Pro Phe Arg Val Asn
      130            135            140
Ser Asp Gly Lys Thr Leu His Gln Asn Arg Tyr Ser Trp Asn Tyr Ala
      145            150            155            160
Ala Asn Val Leu Phe Leu Glu Ser Pro Val Gly Val Gly Phe Ser Tyr
      165            170            175
Ser Asn Lys Ser Thr Glu Tyr Ser Ser Asn Gly Asp Lys Lys Thr Ala
      180            185            190
Ile Asp Asn Tyr Leu Phe Leu Val Asn Trp Leu Glu Arg Phe Pro Glu
      195            200            205
Tyr Lys Asn Arg Asp Phe Tyr Ile Ser Gly Glu Ser Tyr Ala Gly His
      210            215            220
Tyr Val Pro Gln Leu Ala His Thr Ile Leu Tyr His Asn Lys Lys Ala
      225            230            235            240

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tacaacaata aactctataa caacaccatt attaacctca aaggcatttc tatagggaat 780
gcttgggattg atgatgcgac gaatttaaag gggatatatg ataacttggtg gactcatgct 840
ttaaactcag atcaaaactca tgagttgatt gagaagtact gtgacttcac taaagaaaat 900
gtttcagcaa tttgtaacaa tgcaactgat aaggccttcg ttgagacagg aaagatagac 960
atctataaca tccatgcgcc attgtgtcat gactcttctc tgaaaaatgg ttctagtact 1020
ggttacgtaa gcaatgattt tgacccttgt tctgattact atgttactgc ctatctaaat 1080
agaccagaag ttcaaaaggc tcttcatgca aaacctacaa attggacca ttgcactcat 1140
cttcttacta cctggaaaga cagtccagct accgtctac ccaccgtcaa gtatctcatt 1200
gatagcggca ttaaattatg gatatacagt ggtgatacag atgtagtggg tccaaccaca 1260
tcttcaagat atttaacaa cacccttaaa cttccaatca actctgcttg gcgtccgtgg 1320
tattctggaa aagagattgg agggatgtt gtgggataca aaggattgac atttgttaca 1380
gtgagaggag caggacatct tgtccaagc tggcaacctg aacgtgcttt gactttgatc 1440
tcatcattcc tctatggaat cctgccttct ggttcaccgt cgaattaa 1488

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&lt;210&gt; SEQ ID NO 15

&lt;211&gt; LENGTH: 495

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Medicago truncatula

&lt;400&gt; SEQUENCE: 15

```

Met Lys Lys Val Ser Leu Tyr Ala Cys Leu Leu Leu Asn Leu Ser Leu
  1             5             10             15

Leu Val Ile Phe Pro Tyr Ser Lys Ala Ser Gln Ala Asp Lys Phe Asn
      20             25             30

Glu Phe Ile Leu Ser Arg Lys Ser Gln Asn Pro Pro Lys Thr Leu Ser
      35             40             45

Trp Glu Glu Gly Asp Ala Leu Lys Thr His Ser Phe Ser Ala Ala Tyr
      50             55             60

Val Ala Pro Pro Gln Glu Glu Leu Arg Leu Ala Asp Lys Ile Val Thr
      65             70             75             80

Leu Pro Gly Gln Pro Tyr Gly Val Asn Phe Asp Gln Tyr Ser Gly Tyr
      85             90             95

Val Thr Val Asp Pro Glu Ala Gly Arg Glu Leu Phe Tyr Tyr Phe Val
      100            105            110

Glu Ser Pro His Asn Ser Tyr Thr Lys Pro Leu Ile Leu Trp Leu Asn
      115            120            125

Gly Gly Pro Gly Cys Ser Ser Leu Gly Tyr Gly Ala Phe Glu Glu Leu
      130            135            140

Gly Pro Phe Arg Val Asn Ser Asp Gly Lys Thr Leu Tyr Arg Asn Pro
      145            150            155            160

Tyr Ala Trp Asn Glu Val Ala Asn Val Leu Phe Leu Glu Ser Pro Ala
      165            170            175

Gly Val Gly Phe Ser Tyr Ser Asn Thr Ser Ser Asp Tyr Asp Asn Ser
      180            185            190

Gly Asp Lys Ser Thr Ala Lys Asp Ala Tyr Val Phe Leu Ile Asn Trp
      195            200            205

Leu Glu Arg Phe Pro Gln Tyr Lys Thr Arg Asp Phe Tyr Ile Thr Gly
      210            215            220

Glu Ser Tyr Ala Gly His Tyr Val Pro Gln Leu Ala Ser Thr Ile Leu
      225            230            235            240

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Tyr Asn Asn Lys Leu Tyr Asn Asn Thr Ile Ile Asn Leu Lys Gly Ile  
                   245                  250                  255  
 Ser Ile Gly Asn Ala Trp Ile Asp Asp Ala Thr Asn Leu Lys Gly Ile  
                   260                  265                  270  
 Tyr Asp Asn Leu Trp Thr His Ala Leu Asn Ser Asp Gln Thr His Glu  
                   275                  280                  285  
 Leu Ile Glu Lys Tyr Cys Asp Phe Thr Lys Glu Asn Val Ser Ala Ile  
                   290                  295                  300  
 Cys Asn Asn Ala Thr Asp Lys Ala Phe Val Glu Thr Gly Lys Ile Asp  
                   305                  310                  315                  320  
 Ile Tyr Asn Ile His Ala Pro Leu Cys His Asp Ser Ser Leu Lys Asn  
                   325                  330                  335  
 Gly Ser Ser Thr Gly Tyr Val Ser Asn Asp Phe Asp Pro Cys Ser Asp  
                   340                  345                  350  
 Tyr Tyr Val Thr Ala Tyr Leu Asn Arg Pro Glu Val Gln Lys Ala Leu  
                   355                  360                  365  
 His Ala Lys Pro Thr Asn Trp Thr His Cys Thr His Leu Leu Thr Thr  
                   370                  375                  380  
 Trp Lys Asp Ser Pro Ala Thr Val Leu Pro Thr Val Lys Tyr Leu Ile  
                   385                  390                  395                  400  
 Asp Ser Gly Ile Lys Leu Trp Ile Tyr Ser Gly Asp Thr Asp Val Val  
                   405                  410                  415  
 Val Pro Thr Thr Ser Ser Arg Tyr Leu Ile Asn Thr Leu Lys Leu Pro  
                   420                  425                  430  
 Ile Asn Ser Ala Trp Arg Pro Trp Tyr Ser Gly Lys Glu Ile Gly Gly  
                   435                  440                  445  
 Tyr Val Val Gly Tyr Lys Gly Leu Thr Phe Val Thr Val Arg Gly Ala  
                   450                  455                  460  
 Gly His Leu Val Pro Ser Trp Gln Pro Glu Arg Ala Leu Thr Leu Ile  
                   465                  470                  475                  480  
 Ser Ser Phe Leu Tyr Gly Ile Leu Pro Ser Gly Ser Pro Ser Asn  
                   485                  490                  495

&lt;210&gt; SEQ ID NO 16

&lt;211&gt; LENGTH: 1278

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Arabidopsis thaliana

&lt;400&gt; SEQUENCE: 16

```

atgatcaagg cacttcagg gcaaccgcaa gtaggattct cacagttttc gggttatgtg      60
actgtgaacg agtcacatgg tcgatcactt ttctactggc ttacagagtc cccttcttct      120
tctcacacca aaccattctt tctttggctc aatggaggac cgggttgctc atcgattggt      180
tatggagctt cggaggaaat tggaccgttt cgatcaata aaaccggttc taatctctat      240
ctcaacaagt ttactgtgaa cacagaagcg aatattttgt ttcttgaatc gccggctgga      300
gttggttttt cgtacactaa cacaagctct gatcttaaag attctgggga cgaacggact      360
gctcaggaaa atttgatatt tctaattaaa tggatgtcga gatttctca gtaccaatat      420
agagatttct acattgttgg tgaagctac gctggctcatt atgttctca gcttgccaaa      480
aagatccatc tctacaacaa agctttcaac aatacaccca tcattaacct caaaggattc      540
atggtgggaa atggagatat ggacaagcat tacgacagat taggagccgc catgtatgcg      600
tggtcacacg caatgatctc tgacaaaact tacaagtcta tcctcaaaca ctgcagcttc      660

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actgcggata aaacctcgga caagtgcaat tgggcactct acttcgccta cagagagttt 720
ggcaaagtca atgggtacag catctactca ccctcatgtg tacatcaaac caaccagacc 780
aagttcctgc atggacggct tttggtagag gaatacgagt acgacccttg taccgaaagc 840
tacgctgaga tatattacaa cgtctctgat gtgcaacgag ctatgcacgc taatcttacc 900
tccattcctt ataagtggaac attgtgcaat atggttgtga ataacaactg gaaagattcc 960
gagttttcaa tgttgccgat atacaaggaa ctcaactgccg ctggtttgag gatctgggtc 1020
tttagtggcg atacagacgc agtggttcca gtgactggga ctgcacttgc cctcagtaaa 1080
ctcaatcttc cggtgaaaac tccttggtac ccttggtact ccgaaaagca ggtgggagga 1140
tggacagagg tatatgaggg gcttaccttt gcgacgataa gaggggcggg ccacgaagtg 1200
ccggtgttgc aacccgagcg tgcctcact cttttaagat cgttcttggc cggcaaagag 1260
cttccaagat cttattag 1278

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&lt;210&gt; SEQ ID NO 17

&lt;211&gt; LENGTH: 425

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Arabidopsis thaliana

&lt;400&gt; SEQUENCE: 17

```

Met Ile Lys Ala Leu Pro Gly Gln Pro Gln Val Gly Phe Ser Gln Phe
  1             5             10             15
Ser Gly Tyr Val Thr Val Asn Glu Ser His Gly Arg Ser Leu Phe Tyr
          20             25             30
Trp Leu Thr Glu Ser Pro Ser Ser Ser His Thr Lys Pro Leu Leu Leu
          35             40             45
Trp Leu Asn Gly Gly Pro Gly Cys Ser Ser Ile Gly Tyr Gly Ala Ser
          50             55             60
Glu Glu Ile Gly Pro Phe Arg Ile Asn Lys Thr Gly Ser Asn Leu Tyr
          65             70             75             80
Leu Asn Lys Phe Thr Trp Asn Thr Glu Ala Asn Ile Leu Phe Leu Glu
          85             90             95
Ser Pro Ala Gly Val Gly Phe Ser Tyr Thr Asn Thr Ser Ser Asp Leu
          100            105            110
Lys Asp Ser Gly Asp Glu Arg Thr Ala Gln Glu Asn Leu Ile Phe Leu
          115            120            125
Ile Lys Trp Met Ser Arg Phe Pro Gln Tyr Gln Tyr Arg Asp Phe Tyr
          130            135            140
Ile Val Gly Glu Ser Tyr Ala Gly His Tyr Val Pro Gln Leu Ala Lys
          145            150            155            160
Lys Ile His Leu Tyr Asn Lys Ala Phe Asn Asn Thr Pro Ile Ile Asn
          165            170            175
Leu Lys Gly Phe Met Val Gly Asn Gly Asp Met Asp Lys His Tyr Asp
          180            185            190
Arg Leu Gly Ala Ala Met Tyr Ala Trp Ser His Ala Met Ile Ser Asp
          195            200            205
Lys Thr Tyr Lys Ser Ile Leu Lys His Cys Ser Phe Thr Ala Asp Lys
          210            215            220
Thr Ser Asp Lys Cys Asn Trp Ala Leu Tyr Phe Ala Tyr Arg Glu Phe
          225            230            235            240
Gly Lys Val Asn Gly Tyr Ser Ile Tyr Ser Pro Ser Cys Val His Gln
          245            250            255
Thr Asn Gln Thr Lys Phe Leu His Gly Arg Leu Leu Val Glu Glu Tyr

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260	265	270
Glu Tyr Asp Pro Cys Thr	Glu Ser Tyr Ala Glu Ile	Tyr Tyr Asn Arg
275	280	285
Pro Asp Val Gln Arg Ala Met	His Ala Asn Leu Thr	Ser Ile Pro Tyr
290	295	300
Lys Trp Thr Leu Cys Asn Met	Val Val Asn Asn Asn	Trp Lys Asp Ser
305	310	315
Glu Phe Ser Met Leu Pro Ile	Tyr Lys Glu Leu Thr	Ala Ala Gly Leu
325	330	335
Arg Ile Trp Val Phe Ser Gly	Asp Thr Asp Ala Val	Val Pro Val Thr
340	345	350
Gly Thr Arg Leu Ala Leu Ser	Lys Leu Asn Leu Pro	Val Lys Thr Pro
355	360	365
Trp Tyr Pro Trp Tyr Ser	Glu Lys Gln Val Gly	Gly Trp Thr Glu Val
370	375	380
Tyr Glu Gly Leu Thr Phe Ala	Thr Ile Arg Gly Ala	Gly His Glu Val
385	390	395
Pro Val Leu Gln Pro Glu Arg	Ala Leu Thr Leu Arg	Ser Phe Leu
405	410	415
Ala Gly Lys Glu Leu Pro Arg	Ser Tyr	
420	425	

&lt;210&gt; SEQ ID NO 18

&lt;211&gt; LENGTH: 1422

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Arabidopsis thaliana

&lt;400&gt; SEQUENCE: 18

atggcaatgg caaaactgcg aattttcacc actcttatgg ccatactcgt aatgacatct	60
caaggaagga ttccaacaga aggaggagag aaagaagcag aggctgacag aattacgtca	120
cttccaggtc agcctaactg cactgtcgag cagttttccg gctacgtcac cgtcgataaa	180
ctctccggaa gatcactctt ttattggctc actgaagctt ctgacctccc tctctccaaa	240
cctctcgtaa tttggctcaa cggaggaccg ggatgttcgt cggtagcgta cggtagcgctg	300
gaggagattg gaccattcag gataagcaaa ggtggttccg gtttgatatc caacaagttc	360
gcatggaaact caatctccaa tctcttggtc ctggaagctc ccgcccgcgt cggtctctct	420
tactactaac gctcctccga tctcttcaac accggtgata gccgtaccgc caaagattca	480
cttcagtttc ttattcaatg gcttcaccgg ttcccgagat acaaccaccg ggaaatctac	540
atcaccggcg agagttaacg cggacattac gtctctcagc tggccaaaga gatcatgaat	600
tacaacaaac gatcaaagaa tccgttaaat ctcaaaggaa tcatggttgg aaacgcggtg	660
acggacaatc actatgataa cctaggaacg gtttcgtatt ggtggagcca cgcgatgac	720
tctgatcgga cgtatcatca gttgataagc acttgcgatt ttagtcgtca gaaggaatct	780
gatgaatgag aaacccttta ttcttaacgt atggagcagg agtttggtta cattgatcag	840
tacaacatct atgcgccgcc gtgtaacaag tcaagcgacg gtggtggtag ctacaatggt	900
tcttccggcc gccggagtat gcggttctc cacttctccc actccgtatt gagggaaatt	960
tccggttatg atccatgtac cgagagatat gcagagatct attataaccg gcctgatgtt	1020
cagaaagctc ttcacgccaa caccaccaag attccgtata aatggacagc ttgcagtgag	1080
gtgctaaacc ggaattggaa cgacacagat tcaacggttc tccctatata ccgggaaatg	1140
attgccggcg gaattagagt ttgggttttc agtggtgacg tcgattcagt tgtaccagt	1200

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acagctacta gataactcact agcaagactt agtttgagta ccaaacttcc ttggtatcct 1260
tggtatgtca agaaacaggt tggaggatgg acggaagtgt atgaaggact aacgttcgtg 1320
acggttagag gagcagggtca cgagggtgcca ttgttcaagc cactgtctgc ttttgagctt 1380
ttaaagtatt tcttgagagg caagccactt ccaaaggctt aa 1422

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<210> SEQ ID NO 19
<211> LENGTH: 473
<212> TYPE: PRT
<213> ORGANISM: Arabidopsis thaliana

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<400> SEQUENCE: 19

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Met Ala Met Ala Lys Leu Ala Ile Phe Thr Thr Leu Met Ala Ile Leu
  1             5             10             15
Val Met Thr Ser Gln Gly Arg Ile Pro Thr Glu Gly Gly Glu Lys Glu
      20             25             30
Ala Glu Ala Asp Arg Ile Thr Ser Leu Pro Gly Gln Pro Asn Val Thr
      35             40             45
Phe Glu Gln Phe Ser Gly Tyr Val Thr Val Asp Lys Leu Ser Gly Arg
      50             55             60
Ser Leu Phe Tyr Trp Leu Thr Glu Ala Ser Asp Leu Pro Leu Ser Lys
      65             70             75             80
Pro Leu Val Ile Trp Leu Asn Gly Gly Pro Gly Cys Ser Ser Val Ala
      85             90             95
Tyr Gly Ala Ser Glu Glu Ile Gly Pro Phe Arg Ile Ser Lys Gly Gly
      100            105            110
Ser Gly Leu Tyr Leu Asn Lys Phe Ala Trp Asn Ser Ile Ser Asn Leu
      115            120            125
Leu Phe Leu Glu Ala Pro Ala Gly Val Gly Phe Ser Tyr Thr Asn Arg
      130            135            140
Ser Ser Asp Leu Phe Asn Thr Gly Asp Arg Arg Thr Ala Lys Asp Ser
      145            150            155            160
Leu Gln Phe Leu Ile Gln Trp Leu His Arg Phe Pro Arg Tyr Asn His
      165            170            175
Arg Glu Ile Tyr Ile Thr Gly Glu Ser Tyr Ala Gly His Tyr Val Pro
      180            185            190
Gln Leu Ala Lys Glu Ile Met Asn Tyr Asn Lys Arg Ser Lys Asn Pro
      195            200            205
Leu Asn Leu Lys Gly Ile Met Val Gly Asn Ala Val Thr Asp Asn His
      210            215            220
Tyr Asp Asn Leu Gly Thr Val Ser Tyr Trp Trp Ser His Ala Met Ile
      225            230            235            240
Ser Asp Arg Thr Tyr His Gln Leu Ile Ser Thr Cys Asp Phe Ser Arg
      245            250            255
Gln Lys Glu Ser Asp Glu Cys Glu Thr Leu Tyr Ser Tyr Ala Met Glu
      260            265            270
Gln Glu Phe Gly Asn Ile Asp Gln Tyr Asn Ile Tyr Ala Pro Pro Cys
      275            280            285
Asn Lys Ser Ser Asp Gly Gly Gly Ser Tyr Asn Gly Ser Ser Gly Arg
      290            295            300
Arg Ser Met Arg Leu Pro His Leu Pro His Ser Val Leu Arg Lys Ile
      305            310            315            320
Ser Gly Tyr Asp Pro Cys Thr Glu Arg Tyr Ala Glu Ile Tyr Tyr Asn
      325            330            335

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Arg Pro Asp Val Gln Lys Ala Leu His Ala Asn Thr Thr Lys Ile Pro  
                   340                  345                  350

Tyr Lys Trp Thr Ala Cys Ser Glu Val Leu Asn Arg Asn Trp Asn Asp  
           355                  360                  365

Thr Asp Ser Thr Val Leu Pro Ile Tyr Arg Glu Met Ile Ala Gly Gly  
       370                  375                  380

Ile Arg Val Trp Val Phe Ser Gly Asp Val Asp Ser Val Val Pro Val  
   385                  390                  395                  400

Thr Ala Thr Arg Tyr Ser Leu Ala Arg Leu Ser Leu Ser Thr Lys Leu  
                   405                  410                  415

Pro Trp Tyr Pro Trp Tyr Val Lys Lys Gln Val Gly Gly Trp Thr Glu  
       420                  425                  430

Val Tyr Glu Gly Leu Thr Phe Val Thr Val Arg Gly Ala Gly His Glu  
       435                  440                  445

Val Pro Leu Phe Lys Pro Arg Ala Ala Phe Glu Leu Phe Lys Tyr Phe  
       450                  455                  460

Leu Arg Gly Lys Pro Leu Pro Lys Ala  
   465                  470

&lt;210&gt; SEQ ID NO 20

&lt;211&gt; LENGTH: 1359

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Arabidopsis thaliana

&lt;400&gt; SEQUENCE: 20

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atggctcgac tcttctctct cttcttcttc ttccttattc tactccatta cgttcttgt      60
tccagacacg aacaagaaaa agaccgaatc ttccaccttc ccggtgaacc aaacgatgtc      120
tcttctctct acttctctgg ttacattacc gtcaacgagt cagcaggaag agcactattc      180
tactggctca ctgagtcacc accgagtga aacctgagt ctaagcctct tgtcctctgg      240
ctcaacggtg gacctggttg ttctccgta gcttacggtg ccgctgaaga aatcggacct      300
tttagaatca atctgatgg caaaactctt taccacaatc cttactcttg gaacaaattg      360
gcgaatttgc tcttcttga atctctgct ggtgttggtt tctcgtattc gaatactacc      420
tccgatttgt atactgcgg agatcagaga actgcggaag atgcttatgt gtttcttgtg      480
aatgggttgg agaggtttcc tcaatacaaa cacagagagt tctacattgc tggagaaagc      540
tatgcaggtc attatgttcc tcagttgtca cagattgttt atgagaaacg caatccagct      600
atcaacttta aaggcttcat tgttggaat gctgtgattg atgactacca tgattacgtg      660
ggtttatttg aatattggtg ggtcatggg ttgatatctg atctcactta ccacaactta      720
cggatcacgt gtgaatttgg atcatccgag caccgcctct ctaaatgcac caaggccatg      780
gaagctgcag acttgagca aggcaatatt gatccttata gcatttacac tgtcacttgt      840
aaaaaggagg ctgcagctct taggtctcgc ttctcgagag ttctgtatcc atggatgtgg      900
agagcctatg acccttgcac agagaaatac tccggcatgt atttcaattc tccggaggtt      960
caaaaggcta tgcattgtaa tataacagga ctagcttata catggaaagg gtgcagtgac     1020
atcgttgagg agaaatgggc agattctcct ctgtctatgc ttccaatcta caaagaactc     1080
atcgccgcag gtctcaggat atgggttttc agcggagaca ctgattcagt gggtccattt     1140
actggaacac gatactccat tagagccctc aagttacaac cactctccaa atgggtaccct     1200
tggaacgatg atggacaggt tgggtgatgg agccaagttt acaaagggct gactctgggtg     1260
acaatacatg gagcaggaca tgaggtacct cttttccgcc ctctctgagc ttttcttctt     1320

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tttcagtcgt ttctcgacaa caagccattg ccaatgtaa

1359

&lt;210&gt; SEQ ID NO 21

&lt;211&gt; LENGTH: 452

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Arabidopsis thaliana

&lt;400&gt; SEQUENCE: 21

Met Ala Arg Leu Leu Leu Phe Phe Phe Phe Leu Ile Leu Leu His  
 1 5 10 15  
 Tyr Ala Ser Cys Ser Arg His Glu Gln Glu Lys Asp Arg Ile Phe His  
 20 25 30  
 Leu Pro Gly Glu Pro Asn Asp Val Ser Phe Ser His Phe Ser Gly Tyr  
 35 40 45  
 Ile Thr Val Asn Glu Ser Ala Gly Arg Ala Leu Phe Tyr Trp Leu Thr  
 50 55 60  
 Glu Ser Pro Pro Ser Glu Asn Pro Glu Ser Lys Pro Leu Val Leu Trp  
 65 70 75 80  
 Leu Asn Gly Gly Pro Gly Cys Ser Ser Val Ala Tyr Gly Ala Ala Glu  
 85 90 95  
 Glu Ile Gly Pro Phe Arg Ile Asn Pro Asp Gly Lys Thr Leu Tyr His  
 100 105 110  
 Asn Pro Tyr Ser Trp Asn Lys Leu Ala Asn Leu Leu Phe Leu Glu Ser  
 115 120 125  
 Pro Ala Gly Val Gly Phe Ser Tyr Ser Asn Thr Thr Ser Asp Leu Tyr  
 130 135 140  
 Thr Ala Gly Asp Gln Arg Thr Ala Glu Asp Ala Tyr Val Phe Leu Val  
 145 150 155 160  
 Lys Trp Phe Glu Arg Phe Pro Gln Tyr Lys His Arg Glu Phe Tyr Ile  
 165 170 175  
 Ala Gly Glu Ser Tyr Ala Gly His Tyr Val Pro Gln Leu Ser Gln Ile  
 180 185 190  
 Val Tyr Glu Lys Arg Asn Pro Ala Ile Asn Phe Lys Gly Phe Ile Val  
 195 200 205  
 Gly Asn Ala Val Ile Asp Asp Tyr His Asp Tyr Val Gly Leu Phe Glu  
 210 215 220  
 Tyr Trp Trp Ala His Gly Leu Ile Ser Asp Leu Thr Tyr His Asn Leu  
 225 230 235 240  
 Arg Ile Thr Cys Glu Phe Gly Ser Ser Glu His Pro Ser Ser Lys Cys  
 245 250 255  
 Thr Lys Ala Met Glu Ala Ala Asp Leu Glu Gln Gly Asn Ile Asp Pro  
 260 265 270  
 Tyr Ser Ile Tyr Thr Val Thr Cys Lys Lys Glu Ala Ala Ala Leu Arg  
 275 280 285  
 Ser Arg Phe Ser Arg Val Arg His Pro Trp Met Trp Arg Ala Tyr Asp  
 290 295 300  
 Pro Cys Thr Glu Lys Tyr Ser Gly Met Tyr Phe Asn Ser Pro Glu Val  
 305 310 315 320  
 Gln Lys Ala Met His Ala Asn Ile Thr Gly Leu Ala Tyr Pro Trp Lys  
 325 330 335  
 Gly Cys Ser Asp Ile Val Gly Glu Lys Trp Ala Asp Ser Pro Leu Ser  
 340 345 350  
 Met Leu Pro Ile Tyr Lys Glu Leu Ile Ala Ala Gly Leu Arg Ile Trp  
 355 360 365



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Val Phe Ser Gly Asp Thr Asp Ser Val Val Pro Ile Thr Gly Thr Arg  
 370 375 380

Tyr Ser Ile Arg Ala Leu Lys Leu Gln Pro Leu Ser Lys Trp Tyr Pro  
 385 390 395 400

Trp Asn Asp Asp Gly Gln Val Gly Gly Trp Ser Gln Val Tyr Lys Gly  
 405 410 415

Leu Thr Leu Val Thr Ile His Gly Ala Gly His Glu Val Pro Leu Phe  
 420 425 430

Arg Pro Arg Arg Ala Phe Leu Leu Phe Gln Ser Phe Leu Asp Asn Lys  
 435 440 445

Pro Leu Pro Met  
 450

&lt;210&gt; SEQ ID NO 22

&lt;211&gt; LENGTH: 1380

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Arabidopsis thaliana

&lt;400&gt; SEQUENCE: 22

```

atggattact ctttccttct aatcattctc ttactcacia tctctacttc atgttgtgct      60
gtccttctct cttatgtgga agaacaattg agagacagaa tcagtaactt acctggacaa      120
cctagtaatg tcgatttttag acagtactca ggctatgtca ctgtgcatga agaactgtga      180
agagctttgt tctactgggt ggctgagctc ccggtggccc gtgacccaaa gtctagacct      240
ttggttctgt ggctcaatgg tggccctggt tgttctcttg ttgcttatgg tgetgctgaa      300
gaaattggac cttttctgtg tggttctgat ggcaagactc ttcattccaa actttatgct      360
tggaataaat tggcaaaact gctattcttg gagtctccag ctggagtggg tttctcatat      420
tcaaacacaa cttcagatct ttacacaacc ggtgatcaga gaacagctga ggattcgtac      480
atatttcttg tcaactgggt tgagagggtt ccacaataca agcataggga gttttacatt      540
gttgagaaaa gctatgcagg tcattttggt cctcaactgt ctaaaactgt ccatgaaagg      600
aacaagggct tcaagaaccc ggctataaac ctcaaagggt ttatgggtggg aaatgctggt      660
acagatgact atcatgatta tataggaaca ttgaaatact ggtggaatca cggcttcata      720
tccgattcca cgtatcacca actaaagacc gcgtgctact cagtatcctc tcagcactct      780
tcaatgcagt gtatgggtggc tctgagaaat gccgaattag agcaaggaaa tatcgatcca      840
tatagcattt tcacaaaacc ttgcaacagt actgtggcac ttaagagatt cttaaagggt      900
cgctacccat ggatgtcaag agcttatgat ccttgtacag agagatatcc gaatgtgtat      960
tttaaccgct tggacgttca gaaggctctc caccgaaatg tcaactcgtt atcttaccct      1020
tggaagcat gcagtgcacat tgttaggaagc tattgggacg attctcctct gtctatgctt      1080
cctatataca aagaattgat tactgcaggt ctcaaaatat gggctcttcag tggggatata      1140
gatgctgttg ttcctataac cgctacccga tactctgtag atgcactgaa gctagcaacc      1200
atcacgaact ggtaccctg gtacgacctt ggcaaggtag gtgggtggag tcaagtttac      1260
aaaggactta cattagtgc agtagcagga gctggctcatg aagtgcctct acaccgtccc      1320
cggcaagcct ttattctttt cagatccttt ttagagagca aaccaatgcc tatgacttga      1380

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&lt;210&gt; SEQ ID NO 23

&lt;211&gt; LENGTH: 459

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Arabidopsis thaliana

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&lt;400&gt; SEQUENCE: 23

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Met Asp Tyr Ser Phe Leu Leu Ile Ile Leu Leu Leu Thr Ile Ser Thr
 1             5             10             15

Ser Cys Cys Ala Ala Pro Ser Ser Tyr Val Glu Glu Gln Leu Arg Asp
      20             25             30

Arg Ile Ser Asn Leu Pro Gly Gln Pro Ser Asn Val Asp Phe Arg Gln
      35             40             45

Tyr Ser Gly Tyr Val Thr Val His Glu Glu Arg Gly Arg Ala Leu Phe
 50             55             60

Tyr Trp Leu Val Glu Ser Pro Leu Ala Arg Asp Pro Lys Ser Arg Pro
 65             70             75             80

Leu Val Leu Trp Leu Asn Gly Gly Pro Gly Cys Ser Ser Val Ala Tyr
      85             90             95

Gly Ala Ala Glu Glu Ile Gly Pro Phe Arg Val Gly Ser Asp Gly Lys
 100             105             110

Thr Leu His Ser Lys Leu Tyr Ala Trp Asn Lys Leu Ala Asn Leu Leu
 115             120             125

Phe Leu Glu Ser Pro Ala Gly Val Gly Phe Ser Tyr Ser Asn Thr Thr
 130             135             140

Ser Asp Leu Tyr Thr Thr Gly Asp Gln Arg Thr Ala Glu Asp Ser Tyr
 145             150             155             160

Ile Phe Leu Val Asn Trp Phe Glu Arg Phe Pro Gln Tyr Lys His Arg
      165             170             175

Glu Phe Tyr Ile Val Gly Glu Ser Tyr Ala Gly His Phe Val Pro Gln
 180             185             190

Leu Ser Lys Leu Val His Glu Arg Asn Lys Gly Phe Lys Asn Pro Ala
 195             200             205

Ile Asn Leu Lys Gly Phe Met Val Gly Asn Ala Val Thr Asp Asp Tyr
 210             215             220

His Asp Tyr Ile Gly Thr Phe Glu Tyr Trp Trp Asn His Gly Leu Ile
 225             230             235             240

Ser Asp Ser Thr Tyr His Gln Leu Lys Thr Ala Cys Tyr Ser Val Ser
      245             250             255

Ser Gln His Pro Ser Met Gln Cys Met Val Ala Leu Arg Asn Ala Glu
 260             265             270

Leu Glu Gln Gly Asn Ile Asp Pro Tyr Ser Ile Phe Thr Lys Pro Cys
 275             280             285

Asn Ser Thr Val Ala Leu Lys Arg Phe Leu Lys Gly Arg Tyr Pro Trp
 290             295             300

Met Ser Arg Ala Tyr Asp Pro Cys Thr Glu Arg Tyr Ser Asn Val Tyr
 305             310             315             320

Phe Asn Arg Leu Asp Val Gln Lys Ala Leu His Ala Asn Val Thr Arg
      325             330             335

Leu Ser Tyr Pro Trp Lys Ala Cys Ser Asp Ile Val Gly Ser Tyr Trp
 340             345             350

Asp Asp Ser Pro Leu Ser Met Leu Pro Ile Tyr Lys Glu Leu Ile Thr
 355             360             365

Ala Gly Leu Lys Ile Trp Val Phe Ser Gly Asp Thr Asp Ala Val Val
 370             375             380

Pro Ile Thr Ala Thr Arg Tyr Ser Val Asp Ala Leu Lys Leu Ala Thr
 385             390             395             400

Ile Thr Asn Trp Tyr Pro Trp Tyr Asp His Gly Lys Val Gly Gly Trp
      405             410             415

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Ser Gln Val Tyr Lys Gly Leu Thr Leu Val Thr Val Ala Gly Ala Gly  
420 425 430

His Glu Val Pro Leu His Arg Pro Arg Gln Ala Phe Ile Leu Phe Arg  
435 440 445

Ser Phe Leu Glu Ser Lys Pro Met Pro Met Thr  
450 455

<210> SEQ ID NO 24

<211> LENGTH: 1473

<212> TYPE: DNA

<213> ORGANISM: *Oryza sativa*

<400> SEQUENCE: 24

```

atggcgccggg cgcgcgtgct cctggccgcc atcctactgg cgctgtcccc tctcccatg    60
tccctctccg cggcgccggg cggcgagggt gacactggca cggccgaggc ggccgcggac    120
cgaatcacgg cctgcggggg gcagccacgg gtcaacttct ccatgtactc cgggtacgtc    180
accgtcgacg cggccgcggg gcgcgcgctc ttctactggc tcatcgaggc cggcgacccg    240
gcgtccgcgc cgctcgtgct ctggctcaac ggcgggcccg ggtgctctc cgttggttac    300
ggcgcgtccg aggagctcgg cgcgttcgg atcaaccccg acgggaggtc gctctacttg    360
aacccttacc cctggaacag agtggccaac atgctgttct tggactcccc cgcggcgctc    420
ggctactcct actccaacac cacctccgat ctgttcactg ctggtgataa caagacagct    480
catgattcat atgctttctt ggtgaattgg ttggaacggt ttccgcagta caagtaccgt    540
gattttctaca tcgcaggcga gagctatgga gggcactatg tccctcagtt gtctcagcta    600
gtgtaccgga ataacaaaga cgttgaaaag cctatcctaa actttaagg ctttatgggt    660
ggaaatgcgg taatcgatga ttaccatgac tacgttgga catttgagta ctggtggaca    720
cacgggctga tatctgatga tacatatcag aagctgcagg tggcctgtga ttttgaatca    780
tctgctcacg catcagaagc atgtaacaag atttatgaag tggctgaggc tgaacaaggg    840
aacattgatg catacagcat ctatacgct acctgtaaaa aaacttcatt tctcaaacgc    900
agggttaataa ggggtaactc gccatggttg cctagaggat atgatccctg cactgaaaag    960
tactctacga agtactacaa cctaccagaa gtgcaaaaag catttcatgc caatgtcact   1020
ggaataccgt atgcctggac cacctgcagt gatgacttgt tttattattg gaaagattca   1080
ccaagggtcca tgcttctcat ttaccgtgag ctgattgcgg ctggtctaag aatatgggtt   1140
ttcagcggcg acgctgattc tgtagtcccc ctactgcga caagatactc cattgatgca   1200
ctctatctac ctactgtcac taactggtat ccttggtatg atgatgagga ggttgctggt   1260
tggtgtcaag tgtatcaagg ttgacactg gtgacgatcc gaggagcagg gcatgaagtt   1320
cctctccatc gtccacggca agccttaaaa ctctttgagc atttctaca agataagccc   1380
atgcctcaac ctgagtatac ggccgagaac ttgacgaacg agagctgcta ctgctactgc   1440
ttagtgctag ctcttgatca gctgaacat tga                                     1473

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<210> SEQ ID NO 25

<211> LENGTH: 490

<212> TYPE: PRT

<213> ORGANISM: *Oryza sativa*

<400> SEQUENCE: 25

Met Ala Ala Ala Ala Val Leu Leu Ala Ala Ile Leu Leu Ala Leu Ser  
1 5 10 15

Pro	Leu	Pro	Met	Ser	Leu	Ser	Ala	Gly	Gly	Gly	Gly	Gly	Gly	Asp	Thr	
			20					25					30			
Gly	Thr	Ala	Glu	Ala	Ala	Ala	Asp	Arg	Ile	Thr	Ala	Leu	Pro	Gly	Gln	
			35				40					45				
Pro	Arg	Val	Asn	Phe	Ser	Met	Tyr	Ser	Gly	Tyr	Val	Thr	Val	Asp	Ala	
			50				55				60					
Ala	Ala	Gly	Arg	Ala	Leu	Phe	Tyr	Trp	Leu	Ile	Glu	Ala	Ala	Asp	Pro	
			65				70			75						
Ala	Ser	Ala	Pro	Leu	Val	Leu	Trp	Leu	Asn	Gly	Gly	Pro	Gly	Cys	Ser	
			85					90								
Ser	Val	Gly	Tyr	Gly	Ala	Ser	Glu	Glu	Leu	Gly	Ala	Phe	Arg	Ile	Asn	
			100				105									
Pro	Asp	Gly	Arg	Ser	Leu	Tyr	Leu	Asn	Pro	Tyr	Pro	Trp	Asn	Arg	Val	
			115				120									
Ala	Asn	Met	Leu	Phe	Leu	Asp	Ser	Pro	Ala	Gly	Val	Gly	Tyr	Ser	Tyr	
			130				135									
Ser	Asn	Thr	Thr	Ser	Asp	Leu	Phe	Thr	Ala	Gly	Asp	Asn	Lys	Thr	Ala	
			145				150									
His	Asp	Ser	Tyr	Ala	Phe	Leu	Val	Asn	Trp	Leu	Glu	Arg	Phe	Pro	Gln	
			165				170									
Tyr	Lys	Tyr	Arg	Asp	Phe	Tyr	Ile	Ala	Gly	Glu	Ser	Tyr	Gly	Gly	His	
			180				185									
Tyr	Val	Pro	Gln	Leu	Ser	Gln	Leu	Val	Tyr	Arg	Asn	Asn	Lys	Asp	Val	
			195				200									
Glu	Lys	Pro	Ile	Leu	Asn	Phe	Lys	Gly	Phe	Met	Val	Gly	Asn	Ala	Val	
			210				215									
Ile	Asp	Asp	Tyr	His	Asp	Tyr	Val	Gly	Thr	Phe	Glu	Tyr	Trp	Trp	Thr	
			225				230									
His	Gly	Leu	Ile	Ser	Asp	Asp	Thr	Tyr	Gln	Lys	Leu	Gln	Val	Ala	Cys	
			245				250									
Asp	Phe	Glu	Ser	Ser	Ala	His	Ala	Ser	Glu	Ala	Cys	Asn	Lys	Ile	Tyr	
			260				265									
Glu	Val	Ala	Glu	Ala	Glu	Gln	Gly	Asn	Ile	Asp	Ala	Tyr	Ser	Ile	Tyr	
			275				280									
Thr	Pro	Thr	Cys	Lys	Lys	Thr	Ser	Phe	Leu	Lys	Arg	Arg	Leu	Ile	Arg	
			290				295									
Gly	Asn	Ser	Pro	Trp	Leu	Pro	Arg	Gly	Tyr	Asp	Pro	Cys	Thr	Glu	Lys	
			305				310									
Tyr	Ser	Thr	Lys	Tyr	Tyr	Asn	Leu	Pro	Glu	Val	Gln	Lys	Ala	Phe	His	
			325				330									
Ala	Asn	Val	Thr	Gly	Ile	Pro	Tyr	Ala	Trp	Thr	Thr	Cys	Ser	Asp	Asp	
			340				345									
Leu	Phe	Tyr	Tyr	Trp	Lys	Asp	Ser	Pro	Arg	Ser	Met	Leu	Pro	Ile	Tyr	
			355				360									
Arg	Glu	Leu	Ile	Ala	Ala	Gly	Leu	Arg	Ile	Trp	Val	Phe	Ser	Gly	Asp	
			370				375									
Ala	Asp	Ser	Val	Val	Pro	Leu	Thr	Ala	Thr	Arg	Tyr	Ser	Ile	Asp	Ala	
			385				390									
Leu	Tyr	Leu	Pro	Thr	Val	Thr	Asn	Trp	Tyr	Pro	Trp	Tyr	Asp	Asp	Glu	
			405				410									
Glu	Val	Ala	Gly	Trp	Cys	Gln	Val	Tyr	Gln	Gly	Leu	Thr	Leu	Val	Thr	
			420				425									
Ile	Arg	Gly	Ala	Gly	His	Glu	Val	Pro	Leu	His	Arg	Pro	Arg	Gln	Ala	

-continued

435	440	445	
Leu Lys Leu Phe Glu His	Phe Leu Gln Asp Lys	Pro Met Pro Gln Pro	
450	455	460	
Glu Tyr Thr Ala Glu Asn	Leu Thr Asn Glu Ser	Cys Tyr Cys Tyr Cys	
465	470	475	480
Leu Val Leu Ala Leu Asp	Gln Pro Glu His		
485	490		
 <210> SEQ ID NO 26			
<211> LENGTH: 1161			
<212> TYPE: DNA			
<213> ORGANISM: Oryza sativa			
 <400> SEQUENCE: 26			
atgtcatgtc ctggatgctc atcgattgcg tatggagcat ctgaagagat tggcccattt	60		
aggattaaga caaacgggac agggctctat ctgaacaagt actcatggaa cagagaggca	120		
aacctcctgt tcttggaaac acctgcccga gttggctttt catactccaa caccacctct	180		
gatctcaaga catctggtga tgagaggaca gctcaagatg cgttgcagtt cttgatcagt	240		
tggatgtccc gcttcccaca gtatcggcac cgggatttct acattgctgg agaaagctat	300		
gctggacatt acgttcccca gttggcaagg aagatcgttg agttcaacaa ggccctacca	360		
tatcctttca tcaacctcaa ggggatcctt gtgggcaatg gggtgactga caactactac	420		
gacaacatcg gcacgggtgac ctactgggtg acgcacgccca tgatctcggc caccacctac	480		
aaggccatca tgtcgtcgtg caacttcacc agcgccaacg tctccaggct ctgcaaccgc	540		
gccatgagct acgccatgaa ccacgagttc ggcgacatcg accagtacag catctacacg	600		
ccgtcctgcg ccgcccgcgc cgccgccaac gccaccggcc gcccgccggc caaggccgcc	660		
gtgctgaggt tcaaggacac cttcctacgg cgccgggtcgt tcggctacga cccctgcacg	720		
gagacatacg ccgagaagta ctacaaccgg ccggatgttc agaaggccat gcatgccaac	780		
atcactggga ttccttacag atggacagcc tgcagtgatg tgctcatcaa gacgtggcga	840		
gattcagagt tctccatgct gccgacttac aagttgctga tgaaggccgg gctgaggata	900		
tgggtgttca gtggcgacac ggattcagtc gttccgggta ctgcaacgag gtttgcgctt	960		
agccatcttg gactgaagac gaagatccgc tggtagccctt ggtactcagc tggacaggtt	1020		
ggaggatggt ctgaggtgta tgaagggtc acatttgctg cagtgaaggg tgctgggcat	1080		
gaggtgccac tgtttcagcc aaggagagca ttcaggatgt ttcagtcgtt cttggcaggg	1140		
gagccattgc caaaatcctg a	1161		

<210> SEQ ID NO 27  
 <211> LENGTH: 386  
 <212> TYPE: PRT  
 <213> ORGANISM: Oryza sativa

<400> SEQUENCE: 27

Met Ser Cys Pro Gly Cys Ser Ser Ile Ala Tyr Gly Ala Ser Glu Glu	
1 5 10 15	
Ile Gly Pro Phe Arg Ile Lys Thr Asn Gly Thr Gly Leu Tyr Leu Asn	
20 25 30	
Lys Tyr Ser Trp Asn Arg Glu Ala Asn Leu Leu Phe Leu Glu Ser Pro	
35 40 45	
Ala Gly Val Gly Phe Ser Tyr Ser Asn Thr Thr Ser Asp Leu Lys Thr	
50 55 60	

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Ser Gly Asp Glu Arg Thr Ala Gln Asp Ala Leu Gln Phe Leu Ile Ser  
 65 70 75 80  
 Trp Met Ser Arg Phe Pro Gln Tyr Arg His Arg Asp Phe Tyr Ile Ala  
 85 90 95  
 Gly Glu Ser Tyr Ala Gly His Tyr Val Pro Gln Leu Ala Arg Lys Ile  
 100 105 110  
 Val Glu Phe Asn Lys Ala Ser Pro Tyr Pro Phe Ile Asn Leu Lys Gly  
 115 120 125  
 Ile Leu Val Gly Asn Gly Val Thr Asp Asn Tyr Tyr Asp Asn Ile Gly  
 130 135 140  
 Thr Val Thr Tyr Trp Trp Thr His Ala Met Ile Ser Asp Thr Thr Tyr  
 145 150 155 160  
 Lys Ala Ile Met Ser Ser Cys Asn Phe Thr Ser Ala Asn Val Ser Arg  
 165 170 175  
 Leu Cys Asn Arg Ala Met Ser Tyr Ala Met Asn His Glu Phe Gly Asp  
 180 185 190  
 Ile Asp Gln Tyr Ser Ile Tyr Thr Pro Ser Cys Ala Ala Ala Ala Ala  
 195 200 205  
 Ala Asn Ala Thr Gly Arg Arg Arg Gly Lys Ala Ala Val Leu Arg Phe  
 210 215 220  
 Lys Asp Thr Phe Leu Arg Arg Arg Ser Phe Gly Tyr Asp Pro Cys Thr  
 225 230 235 240  
 Glu Thr Tyr Ala Glu Lys Tyr Tyr Asn Arg Pro Asp Val Gln Lys Ala  
 245 250 255  
 Met His Ala Asn Ile Thr Gly Ile Pro Tyr Arg Trp Thr Ala Cys Ser  
 260 265 270  
 Asp Val Leu Ile Lys Thr Trp Arg Asp Ser Glu Phe Ser Met Leu Pro  
 275 280 285  
 Thr Tyr Lys Leu Leu Met Lys Ala Gly Leu Arg Ile Trp Val Phe Ser  
 290 295 300  
 Gly Asp Thr Asp Ser Val Val Pro Val Thr Ala Thr Arg Phe Ala Leu  
 305 310 315 320  
 Ser His Leu Gly Leu Lys Thr Lys Ile Arg Trp Tyr Pro Trp Tyr Ser  
 325 330 335  
 Ala Gly Gln Val Gly Gly Trp Ser Glu Val Tyr Glu Gly Leu Thr Phe  
 340 345 350  
 Ala Ser Val Arg Gly Ala Gly His Glu Val Pro Leu Phe Gln Pro Arg  
 355 360 365  
 Arg Ala Phe Arg Met Phe Gln Ser Phe Leu Ala Gly Glu Pro Leu Pro  
 370 375 380  
 Lys Ser  
 385

&lt;210&gt; SEQ ID NO 28

&lt;211&gt; LENGTH: 1458

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Oryza sativa

&lt;400&gt; SEQUENCE: 28

atggccggcg ctaccgctgc cgccgtctcc tctctcttcc tcgcgctcgc gttgctctcg	60
ctctgcgcgc cgccgctggt cggtctgcct cagctggacg cggaggccgc gcggcagcag	120
gaggccgacc gcgtgacgag gctgccgggg caaccgccc tgccggttcgc gcagtagccc	180
gggtacgtga cgggtgaacga gacgcacggc cgcgcgctct tctactggtt cttcgaggcc	240

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accgccgccc	ccgacaagaa	gccccctcgtc	ctctgggtcca	acggcggggcc	tgggtgttcg	300
tctgttgggt	atggagaagc	ggaggagctc	ggtccattct	tggcgagaa	gggcaagccg	360
gagctaaaa	ggaacaagta	ctcgtggaac	aaagaggcca	atctgatgtt	cctggagtcc	420
cctgtgggtg	tcggcttctc	atacactaac	acaagctccg	atctgcagca	gcttggcgac	480
aagatcaccc	ctgatgatgc	ttacatcttc	ctgctcaact	ggttcaagcg	cttcctcag	540
tacaaatctc	acgacttcta	catcgctgga	gagagctacg	ctgggcatta	cgttccacag	600
ctttcggaga	agattttcga	cggcaacaag	caaggcccca	aggagaacta	catcaacttc	660
aagggtttca	tgatagggaa	tgccctgatg	gacgacgaga	cggaccagac	gggcatgatc	720
gactacgcct	gggaccacgc	cgtcatctcg	gaccgggtgt	acgccgacgt	caagaagtac	780
tgcaacttca	gcattggagaa	cgtgaccgac	gcgtgcgaca	gcgcgctcac	cgagtacttc	840
gccgtgtacc	gcctcatcga	catgtacagc	ctctacaccc	ccgtctgcac	cgaggctctg	900
tcgtcggcgg	cgttcggcca	gcgccaggtc	gccgtccacg	gcgccgcccc	aaaaatcttc	960
tccaaatacc	atgggtggta	catgaggccg	gcgggggtacg	atccgtgcac	gtcggatcac	1020
gccgagggtg	acttcaaccg	ggctgacgtg	caggaggcgc	tgcacgcca	cgtgaccaat	1080
atcggttaca	actggacgca	ctgcagcgac	gtgatcgcca	agtgagagaga	tgctcccttc	1140
tcgactctcc	ccatcatccg	taagctcgtc	gccggcggca	tcagggtctg	ggttttcagc	1200
ggtgacaccg	atggaaggat	ccccgtgacg	tcgacgaggc	tcaccctgaa	caagcttggg	1260
ctgaagacgg	tgcaggagtg	gacgccgtgg	tacgaccatc	agcagggttg	aggatggacg	1320
atcctctacg	agggcctgac	gttcgtgacg	atccgcggcg	ccgggcacga	ggttcccttg	1380
cacgcgccga	ggcaggcgct	cagcctcttc	agccacttct	tggtgacaaa	gaagatgcct	1440
ccgacggcgt	tcctctag					1458

&lt;210&gt; SEQ ID NO 29

&lt;211&gt; LENGTH: 485

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Oryza sativa

&lt;400&gt; SEQUENCE: 29

Met	Ala	Gly	Ala	Thr	Ala	Ala	Ala	Val	Ser	Ser	Ser	Phe	Leu	Ala	Leu
1				5					10					15	
Ala	Leu	Leu	Ser	Leu	Cys	Ala	Ala	Ala	Ala	Gly	Gly	Ser	Pro	Gln	Leu
			20					25					30		
Asp	Ala	Glu	Ala	Ala	Arg	Gln	Gln	Glu	Ala	Asp	Arg	Val	Thr	Arg	Leu
			35				40					45			
Pro	Gly	Gln	Pro	Ala	Val	Arg	Phe	Ala	Gln	Tyr	Ala	Gly	Tyr	Val	Thr
			50				55				60				
Val	Asn	Glu	Thr	His	Gly	Arg	Ala	Leu	Phe	Tyr	Trp	Phe	Phe	Glu	Ala
					70					75				80	
Thr	Ala	Ala	Ala	Asp	Lys	Lys	Pro	Leu	Val	Leu	Trp	Leu	Asn	Gly	Gly
				85				90					95		
Pro	Gly	Cys	Ser	Ser	Val	Gly	Tyr	Gly	Glu	Ala	Glu	Glu	Leu	Gly	Pro
			100					105					110		
Phe	Leu	Val	Gln	Lys	Gly	Lys	Pro	Glu	Leu	Lys	Trp	Asn	Lys	Tyr	Ser
			115				120					125			
Trp	Asn	Lys	Glu	Ala	Asn	Leu	Met	Phe	Leu	Glu	Ser	Pro	Val	Gly	Val
			130			135					140				
Gly	Phe	Ser	Tyr	Thr	Asn	Thr	Ser	Ser	Asp	Leu	Gln	Gln	Leu	Gly	Asp
					150					155				160	

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Lys Ile Thr Ala Asp Asp Ala Tyr Ile Phe Leu Leu Asn Trp Phe Lys  
 165 170 175  
 Arg Phe Pro Gln Tyr Lys Ser His Asp Phe Tyr Ile Ala Gly Glu Ser  
 180 185 190  
 Tyr Ala Gly His Tyr Val Pro Gln Leu Ser Glu Lys Ile Phe Asp Gly  
 195 200 205  
 Asn Lys Gln Gly Pro Lys Glu Asn Tyr Ile Asn Phe Lys Gly Phe Met  
 210 215 220  
 Ile Gly Asn Ala Leu Met Asp Asp Glu Thr Asp Gln Thr Gly Met Ile  
 225 230 235 240  
 Asp Tyr Ala Trp Asp His Ala Val Ile Ser Asp Arg Val Tyr Ala Asp  
 245 250 255  
 Val Lys Lys Tyr Cys Asn Phe Ser Met Glu Asn Val Thr Asp Ala Cys  
 260 265 270  
 Asp Ser Ala Leu Thr Glu Tyr Phe Ala Val Tyr Arg Leu Ile Asp Met  
 275 280 285  
 Tyr Ser Leu Tyr Thr Pro Val Cys Thr Glu Val Ser Ser Ser Ala Ala  
 290 295 300  
 Phe Gly Gln Arg Gln Val Ala Val His Gly Ala Ala Pro Lys Ile Phe  
 305 310 315 320  
 Ser Lys Tyr His Gly Trp Tyr Met Arg Pro Ala Gly Tyr Asp Pro Cys  
 325 330 335  
 Thr Ser Asp His Ala Glu Val Tyr Phe Asn Arg Ala Asp Val Gln Glu  
 340 345 350  
 Ala Leu His Ala Asn Val Thr Asn Ile Gly Tyr Asn Trp Thr His Cys  
 355 360 365  
 Ser Asp Val Ile Gly Lys Trp Arg Asp Ala Pro Phe Ser Thr Leu Pro  
 370 375 380  
 Ile Ile Arg Lys Leu Val Ala Gly Gly Ile Arg Val Trp Val Phe Ser  
 385 390 395 400  
 Gly Asp Thr Asp Gly Arg Ile Pro Val Thr Ser Thr Arg Leu Thr Leu  
 405 410 415  
 Asn Lys Leu Gly Leu Lys Thr Val Gln Glu Trp Thr Pro Trp Tyr Asp  
 420 425 430  
 His Gln Gln Val Gly Gly Trp Thr Ile Leu Tyr Glu Gly Leu Thr Phe  
 435 440 445  
 Val Thr Ile Arg Gly Ala Gly His Glu Val Pro Leu His Ala Pro Arg  
 450 455 460  
 Gln Ala Leu Ser Leu Phe Ser His Phe Leu Ala Asp Lys Lys Met Pro  
 465 470 475 480  
 Pro Thr Ala Phe Pro  
 485

&lt;210&gt; SEQ ID NO 30

&lt;211&gt; LENGTH: 1449

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Oryza sativa

&lt;400&gt; SEQUENCE: 30

```

atgaaggttc agacttcgtc accttgettg ctactcctac ttggetctct tgcactggtt    60
acactgacac tgtgtggccc agctgcttct gcacggcctg aaacgggcag cctcgatgca    120
tcagccacgg cgccatgga gttgcaggag ctgcaccgag tgatgtcgct gccggggcag    180
ccggcctact cgccgaatt caggcaatac tccggetatg tcaccactga cgagtacctt    240

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ggcaaggcac tcttctactg gttcttggag gccactgaca agcctgacga gaagccactc 300
gtcttgtggc taaatggagg acctggatgt tcttccattg gggttggaca ggcacaggag 360
ctagggccat ttctggtgaa gaaagatgtg gctgaacttg agctgaatcc atacgcatgg 420
aaccaagttg ccaatttgcg gttcctggac tctcctgctg gtgttgggtt ttcttacacc 480
aacacatcct ttgaaaaaga tccaccagga gacaattcca ccgcatatgg ttcatacact 540
ttcctgatca ggtggttcca gaggttcctc cagcacaaaa tgaaggagtt ctacatagct 600
ggagagagct atgcaggaca ttacgttccc cagcttgcta atgtgattgt ggatcagaac 660
aagattgcac ctaagaaaaa ttatataaac ttgaaaggca tcatgatagg aaatgcttac 720
atggatgggt acacggatgt gctaggaatt gttgattctg catggcatca cgcactcatc 780
tcagacaaac tttacagtga ctttcagaag ttctgcaact tcagtttggg tgatctgtct 840
aaagagtgcg acgctgcaat cgatcagttc aacgctctct acagcatcat agatatctac 900
agcctttaca cccctcgatg cgagctcgga tacccaaact tcaactcgtc gtttgcagca 960
caaatcggac ggaccagcag ccgtatacca atgggctatg atccatgctc gaaaacgtac 1020
gcgactgaat atttcaaccg taaagatgtt cagaaaagctc tgcattgcaa tatccctgga 1080
gcatactccc tttgccataa ttctatcaac cgagcatgga acgactctga catgactgtc 1140
cttccaatcg tcaagaaact cactcaatca gggctccgga tatggattta cagcgcgac 1200
acggacgcaa gaatccctac aacctcaacc aggtacacgc tgaaaaagct tgacctgccc 1260
atcaaagagg actggtcgcc atggttccat cacaagcagg ttggtgggtg gagtgtggtg 1320
ttcgacggac tgacatttgt cagggtgaga ggagccggcc acatggtgcc atccatcatg 1380
ccagagcaag cgcttgagct gttcaagtac ttctgggcca atcagaacct cccatccaag 1440
ccattctag

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&lt;210&gt; SEQ ID NO 31

&lt;211&gt; LENGTH: 482

&lt;212&gt; TYPE: PRT

<213> ORGANISM: *Oryza sativa*

&lt;400&gt; SEQUENCE: 31

```

Met Lys Val Gln Thr Ser Ser Pro Cys Leu Leu Leu Leu Gly Ser
  1             5             10             15
Leu Ala Leu Val Thr Leu Thr Leu Cys Gly Pro Ala Ala Ser Ala Arg
  20             25             30
Pro Glu Thr Gly Ser Leu Asp Ala Ser Ala Thr Ala Ala Met Glu Leu
  35             40             45
Gln Glu Leu Asp Arg Val Met Ser Leu Pro Gly Gln Pro Ala Tyr Ser
  50             55             60
Pro Glu Phe Arg Gln Tyr Ser Gly Tyr Val Thr Thr Asp Glu Tyr Leu
  65             70             75             80
Gly Lys Ala Leu Phe Tyr Trp Phe Leu Glu Ala Thr Asp Lys Pro Asp
  85             90             95
Glu Lys Pro Leu Val Leu Trp Leu Asn Gly Gly Pro Gly Cys Ser Ser
  100            105            110
Ile Gly Phe Gly Gln Ala Gln Glu Leu Gly Pro Phe Leu Val Lys Lys
  115            120            125
Asp Val Ala Glu Leu Glu Leu Asn Pro Tyr Ala Trp Asn Gln Val Ala
  130            135            140
Asn Leu Leu Phe Leu Asp Ser Pro Ala Gly Val Gly Phe Ser Tyr Thr

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145	150	155	160
Asn Thr Ser Phe Gly Lys Asp Pro Pro Gly Asp Asn Ser Thr Ala Tyr	165	170	175
Gly Ser Tyr Thr Phe Leu Ile Arg Trp Phe Gln Arg Phe Pro Gln His	180	185	190
Lys Met Lys Glu Phe Tyr Ile Ala Gly Glu Ser Tyr Ala Gly His Tyr	195	200	205
Val Pro Gln Leu Ala Asn Val Ile Val Asp Gln Asn Lys Ile Ala Pro	210	215	220
Lys Glu Asn Tyr Ile Asn Leu Lys Gly Ile Met Ile Gly Asn Ala Tyr	225	230	235
Met Asp Gly Asp Thr Asp Leu Leu Gly Ile Val Asp Ser Ala Trp His	245	250	255
His Ala Leu Ile Ser Asp Lys Leu Tyr Ser Asp Phe Gln Lys Phe Cys	260	265	270
Asn Phe Ser Leu Val Asp Leu Ser Lys Glu Cys Asn Ala Ala Ile Asp	275	280	285
Gln Phe Asn Ala Leu Tyr Ser Ile Ile Asp Ile Tyr Ser Leu Tyr Thr	290	295	300
Pro Arg Cys Glu Leu Gly Tyr Pro Asn Phe Asn Ser Ser Phe Ala Ala	305	310	315
Gln Ile Gly Arg Thr Ser Ser Arg Ile Pro Met Gly Tyr Asp Pro Cys	325	330	335
Ser Gln Thr Tyr Ala Thr Glu Tyr Phe Asn Arg Lys Asp Val Gln Lys	340	345	350
Ala Leu His Ala Asn Ile Pro Gly Ala Tyr Ser Leu Cys His Asn Ser	355	360	365
Ile Asn Arg Ala Trp Asn Asp Ser Asp Met Thr Val Leu Pro Ile Val	370	375	380
Lys Lys Leu Thr Gln Ser Gly Leu Arg Ile Trp Ile Tyr Ser Gly Asp	385	390	395
Thr Asp Ala Arg Ile Pro Thr Thr Ser Thr Arg Tyr Thr Leu Lys Lys	405	410	415
Leu Gly Leu Pro Ile Lys Glu Asp Trp Ser Pro Trp Phe His His Lys	420	425	430
Gln Val Gly Gly Trp Ser Val Val Phe Asp Gly Leu Thr Phe Val Thr	435	440	445
Val Arg Gly Ala Gly His Met Val Pro Ser Ile Met Pro Glu Gln Ala	450	455	460
Leu Glu Leu Phe Lys Tyr Phe Leu Ala Asn Gln Asn Leu Pro Ser Lys	465	470	475
Pro Phe			

&lt;210&gt; SEQ ID NO 32

&lt;211&gt; LENGTH: 1314

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Oryza sativa

&lt;400&gt; SEQUENCE: 32

atggagttgc aggagctcga ccgcgtgatg tcgctgcccg ggcagccggc ctactcgccg 60

gaattcaggc aatactccgg ctatgtcacc actgacgagt accttgga ggcactcttc 120

tactggttct tggaggccac tgacaagcct gacgagaagc cactcgtctt gtggctaaat 180

ggaggacctg gatgttcttc cattgggttt ggacaggcac aggagctagg gccatttctg 240

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gtgaagaaag atgtggctga acttgagctg aatccatacg catggaacca agttgccaat 300
ttgctgttcc tggactctcc tgetggtgtt gggttttctt acaccaacac atcctttgga 360
aaagatccac caggagacaa ttccaccgca tatggttcat acactttcct gatcagggtg 420
ttccagaggt tccctcagca caaaatgaag gagttctaca tagctggaga gagctatgca 480
ggacattacg tccccagct tgctaattgt attgtggatc agaacaagat tgcacctaaa 540
gaaaattata taaacttgaa aggcacatcg ataggaaatg cttacatgga tggtgacacg 600
gatttgctag gaattgttga ttctgcatgg catcacgcac tcatctcaga caaactttac 660
agtgaacttc agaagttctg caacttcagt ttggttgatc tgtctaaaga gtgcaacgct 720
gcaatcgatc agttcaacgc tctctacagc atcatagata tctacagcct ttacaccct 780
cgatcgagc tcggataccc aaacttcaac tcgtcgtttg cagcacaat cggacggacc 840
agcagccgta taccaatggg ctatgatcca tgctcgcaa cgtacgcgac tgaatatctc 900
aaccgtaaag atgttcagaa agctctgcat gccaatatcc ctggagcata ctccctttgc 960
cataattcta tcaaccgagc atggaacgac tctgacatga ctgtccttcc aatcgtaag 1020
aaactcactc aatcagggtt ccggatatgg atttacagcg gcgacacgga cgcaagaatc 1080
cctacaacct caaccaggta cagctgaaa aagcttggcc tgcccatcaa agaggactgg 1140
tcgccatggt tccatcacia gcaggttggg ggggtggagt tggtgttcga cggactgaca 1200
tttgtcacgg tgagaggagc cgccacatg gtgccatcca tcatgccaga gcaagcgctt 1260
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&lt;210&gt; SEQ ID NO 33

&lt;211&gt; LENGTH: 437

&lt;212&gt; TYPE: PRT

<213> ORGANISM: *Oryza sativa*

&lt;400&gt; SEQUENCE: 33

```

Met Glu Leu Gln Glu Leu Asp Arg Val Met Ser Leu Pro Gly Gln Pro
  1           5           10          15
Ala Tyr Ser Pro Glu Phe Arg Gln Tyr Ser Gly Tyr Val Thr Thr Asp
          20          25          30
Glu Tyr Leu Gly Lys Ala Leu Phe Tyr Trp Phe Leu Glu Ala Thr Asp
          35          40          45
Lys Pro Asp Glu Lys Pro Leu Val Leu Trp Leu Asn Gly Gly Pro Gly
          50          55          60
Cys Ser Ser Ile Gly Phe Gly Gln Ala Gln Glu Leu Gly Pro Phe Leu
          65          70          75          80
Val Lys Lys Asp Val Ala Glu Leu Glu Leu Asn Pro Tyr Ala Trp Asn
          85          90          95
Gln Val Ala Asn Leu Leu Phe Leu Asp Ser Pro Ala Gly Val Gly Phe
          100         105         110
Ser Tyr Thr Asn Thr Ser Phe Gly Lys Asp Pro Pro Gly Asp Asn Ser
          115         120         125
Thr Ala Tyr Gly Ser Tyr Thr Phe Leu Ile Arg Trp Phe Gln Arg Phe
          130         135         140
Pro Gln His Lys Met Lys Glu Phe Tyr Ile Ala Gly Glu Ser Tyr Ala
          145         150         155         160
Gly His Tyr Val Pro Gln Leu Ala Asn Val Ile Val Asp Gln Asn Lys
          165         170         175
Ile Ala Pro Lys Glu Asn Tyr Ile Asn Leu Lys Gly Ile Met Ile Gly

```



UNITED STATES PATENT AND TRADEMARK OFFICE  
**CERTIFICATE OF CORRECTION**

PATENT NO. : 7,601,886 B2  
APPLICATION NO. : 11/198886  
DATED : October 13, 2009  
INVENTOR(S) : John C. Walker et al.

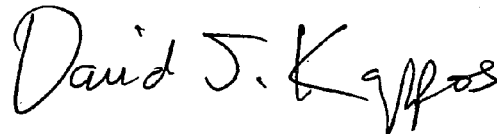
Page 1 of 1

It is certified that error appears in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

In claim 1, column 91, line 47, delete "isloated" and insert --isolated-- therefor.

Signed and Sealed this

Twenty-ninth Day of December, 2009

A handwritten signature in black ink, reading "David J. Kappos". The signature is written in a cursive, flowing style with a large initial 'D' and a stylized 'K'.

David J. Kappos  
*Director of the United States Patent and Trademark Office*

UNITED STATES PATENT AND TRADEMARK OFFICE  
**CERTIFICATE OF CORRECTION**

PATENT NO. : 7,601,886 B2  
APPLICATION NO. : 11/198886  
DATED : October 13, 2009  
INVENTOR(S) : Walker et al.

Page 1 of 1

It is certified that error appears in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

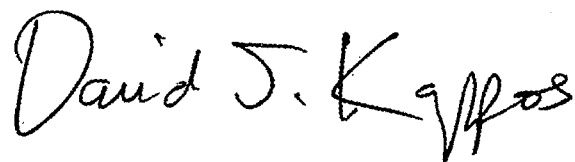
On the Title Page:

The first or sole Notice should read --

Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 212 days.

Signed and Sealed this

Fifth Day of October, 2010

A handwritten signature in black ink, reading "David J. Kappos". The signature is written in a cursive, flowing style with a large initial "D".

David J. Kappos  
*Director of the United States Patent and Trademark Office*